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Lactate Metabolism in Patients with Metastatic Colorectal Cancer[1]

Christopher P. Holroyde,[2] Rita S. Axelrod, Charles L. Skutches, Agnes C. Haff, Pavle Paul, and George A. Reichard

Clinical Oncology Program and Department of Research, Lankenau Hospital, Philadelphia, Pennsylvania 19151

ABSTRACT

The possible effect of tumor glycolysis on whole-body lactate metabolism has not been previously measured in humans. Using an isotope tracer technique, we found increased rates of lactate production in 20 patients with metastatic colorectal cancer compared to 13 control subjects of comparable age and sex (15.6 ± 1.3 μmol/kg/min *versus* 10.4 ± 0.6 μmol/kg/min; $p < 0.01$). The cancer patients were characterized by a moderately increased rate of lactate oxidation ($p < 0.05$), a 2-fold increase in nonoxidative lactate disposal ($p < 0.001$), and an increased percentage of glucose derived from lactate ($p < 0.01$). There was a direct correlation between venous plasma lactate concentrations and lactate production rates. There was also a direct correlation between lactate production rates and the percentage of respiratory CO_2 derived from plasma lactate, the rates of lactate oxidation, the rates of nonoxidative lactate disposal, and the percentage of plasma glucose derived from plasma lactate. These observed results correlated poorly with tumor burden, and no correlation was observed between increased lactate production and the following clinical or biochemical parameters: ambulatory status, histological differentiation, weight change, carcinoembryonic antigen titer, hemoglobin, WBC, presence or absence of hepatic metastases, O_2 consumption, CO_2 production, respiratory quotient, plasma free fatty acids, glucose, and immunoreactive insulin concentrations. This study emphasizes that metabolic heterogeneity with respect to lactate metabolism exists within an apparently homogeneous group of patients with colorectal cancer.

INTRODUCTION

A high rate of glucose utilization with production of lactic acid has long been regarded as a characteristic feature of the neoplastic cell (21). Evidence in support of this assumption is largely derived from *in vitro* studies on human and animal malignant cells or tissue, showing increased rates of glycolysis compared to normal tissue counterparts (1). Kinetic studies of glucose metabolism in normoglycemic cancer patients, however, show only moderate differences in the rates of total glucose turnover and Cori cycle activity when mean values are compared to those of normal subjects (17, 24). On the other hand, total glucose turnover and Cori cycle activity may be strikingly elevated in some cancer patients in whom blood glucose and lactate levels are essentially normal (7, 8, 23). These observations show, indirectly, that tumor glycolysis can lead to increased lactate production and that host-tumor competition for glucose may significantly effect overall fuel economy.

With the exception of isolated case reports in patients with hypoglycemia who were receiving exogenous glucose infusions (12, 13), lactate production has not been directly measured, to our knowledge, in a homogeneous group of cancer patients with a common solid tumor. We have measured rates of lactate production and disposal in 20 patients with metastatic colorectal cancer in an attempt to evaluate the biological significance of altered lactate metabolism and its effect on overall fuel economy of the cancer-bearing host.

MATERIALS AND METHODS

Patients. Twenty patients with histologically proven adenocarcinoma of the colon or rectum were selected for study, and all had documented metastatic disease. Sixteen of 20 patients were partly or completely ambulatory and 4 were hospitalized. None of the patients was completely bedridden and, in all cases, life expectancy was estimated to be >60 days. At the time of the study, no patients were acutely ill, none were febrile, and no chemotherapy or glucocorticoids were administered during the preceding 4 weeks. Thirteen healthy adults of comparable age and sex served as control subjects. Included in the control group were 2 former patients with Duke's Stage A resected colon carcinoma who were presumed to be cured of their disease. The mean weight of the cancer patients was 70.6 kg (range, 48.0 to 95.0 kg) compared to 75.8 kg (range, 60.3 to 96.4 kg) in control subjects. Mean weight change from the time of surgical resection of the primary tumor to the time of study was −7% (range, +6 to −33%). Mean surface area of the cancer patients was 1.82 sq m as compared to 1.88 sq m in control subjects. The ratio of mean weight to mean surface area was 38.8 for the cancer patients compared to 40.3 for the controls. Formal antecedent dietary histories were not obtained.

To every subject, the purpose and potential risks of the isotope tracer procedure were explained, and informed consent was obtained from each.

Metabolic Studies and Methods. Metabolic studies were performed on each subject after an overnight fast of 12 to 14 hr and following a rest period of 30 min in bed before tracer infusion began. Plasma lactate production, oxidation, and conversion to plasma glucose were determined by an isotope tracer technique, using the primed-continuous infusion of 50 to 100 μCi L(+)-[U-^{14}C]lactate over 6 hr. Beginning 1 hr following the initiation of tracer infusion, samples of blood, drawn without stasis, and expired air were obtained at hr intervals. Ultrafiltrates of plasma were prepared using a Millipore filter system containing Pellicon membrane discs with a minimal molecular weight cutoff of 10,000 (Millipore Corp., Bedford, Mass.). Preliminary studies with this system showed complete

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[2] To whom requests for reprints should be addressed.

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recovery of [^{14}C]lactate and nonradioactive lactate added to the plasma. The ultrafiltrates were passed through a column of Amberlite MB-3 (HCO_3^- form) resin as previously described (7), and the glucose-containing eluate was collected. For radioactivity assay, glucose was isolated as gluconic acid (7). Lactate was eluted from the column with 0.1 M NaCl (17). Following addition of carrier lactate to this eluate, radioactivity was determined by oxidation of lactate to acetaldehyde using ceric sulfate, with continuous aeration of the acetaldehyde into a trapping solution of dimedone in an acetate buffer (10). Radioactivity in the bismethone derivative of acetaldehyde represented two-thirds of the total radioactivity of plasma lactate and was corrected accordingly. Limited solubility of the bismethone derivative in the buffer solution (25) generally resulted in the loss of about 15 to 20% of the derivative. Correction for this loss was made on the basis of the theoretical derivative weight calculated from the known quantity of lactate in the oxidized sample.

Respiratory gas samples were obtained simultaneously with each blood sample and analyzed for O_2, CO_2, and $^{14}CO_2$ content by standard methods (9). Immunoreactive insulin was determined by the method of Hales and Randle (5), plasma free fatty acids were determined by the method of Dole and Meinertz (4), plasma lactate was determined by the enzymatic method of Hohorst (6), and plasma glucose was determined using Glucostat reagent (Worthington Biochemical Corp., Freehold, N. J.).

Chart 1 shows time-course changes in the specific activities of plasma lactate, plasma glucose, and respiratory CO_2 in 3 representative studies of this series. During the isotope tracer infusion period in all studies, the plasma lactate concentration and specific activity were constant, indicating that steady state conditions were present. Plasma glucose concentrations were also constant but the specific activities rose gradually, reaching constant values by the fifth and sixth hr in all studies. Respiratory CO_2 specific activities rose throughout each study period but at gradually decreasing rates. Plasma lactate production rates were calculated from the average plasma lactate specific activity and the rate of tracer infusion (14). The percentage of plasma glucose derived from plasma lactate was calculated from the average plasma lactate specific activity and the average of the plasma glucose specific activities at the fifth and sixth hr of tracer infusion (14). The percentage of respiratory CO_2 derived from plasma lactate oxidation and the rate of plasma lactate oxidation were calculated as described (9) with recent modifications (16).

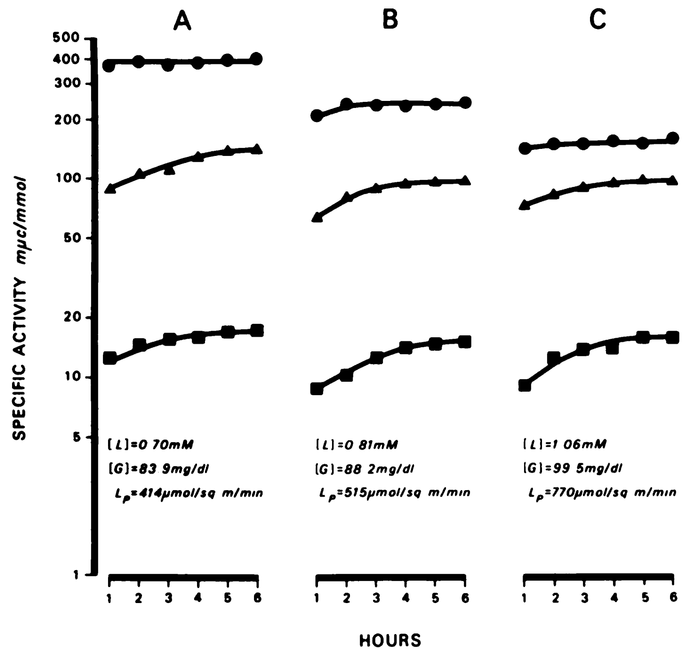

Chart 1. Time-course changes in the specific activities of plasma lactate (●), plasma glucose (▲), and respiratory CO_2 (■) in a typical control subject (*A*), a cancer patient with an intermediate rate of lactate production (*B*), and a cancer patient with a high rate of lactate production (*C*). For comparative purposes, the specific activity data were normalized to the same [^{14}C]lactate infusion rate per kg body weight. Steady state plasma lactate ([*L*]), and plasma glucose ([G]) concentrations and lactate production rates (L_p) for each study are shown. *mμc*, nCi.

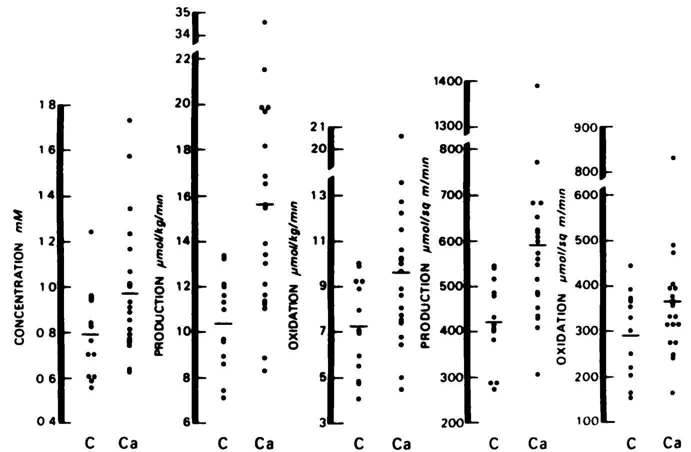

Chart 2. Comparison of plasma lactate concentration and production and oxidation rates in control subjects (*C*) and in patients with metastatic colorectal cancer (*Ca*). *Horizontal bar*, mean of each set of values.

RESULTS

Plasma lactate concentrations and rates of production and oxidation for control subjects and cancer patients are shown in Chart 2. The mean (±S.E.) fasting-venous plasma lactate concentration for the cancer patients was 0.97 ± 0.07 mM compared to 0.79 ± 0.05 mM in the controls ($p < 0.05$). Lactate production rates were significantly increased in the cancer patients (15.6 ± 1.3 *versus* 10.4 ± 0.6 μmol/kg/min; $p < 0.01$). This difference was also observed when production rates were calculated on the basis of surface area rather than body weight (590 ± 49 *versus* 418 ± 26 μmol/sq m/min; $p < 0.01$). (Calculations were made on the basis of body weight and surface area to obviate bias due to changing patient weight not reflected by comparable changes in surface area.) Increased rates of lactate oxidation were observed in patients (9.59 ± 0.80 compared to 7.23 ± 0.58 μmol/kg/min in controls; $p < 0.05$). Lactate oxidation was increased over controls when calculated on the basis of surface area, but the difference was not statistically significant (362 ± 30 *versus* 290 ± 26 μmol/sq m/min). As shown in Chart 2, considerable overlap in values for plasma lactate concentration, production, and oxidation were found between patients and controls, with roughly 50% of patients falling within the normal ranges.

There was a direct correlation between venous plasma lactate concentration and the rate of lactate production, ex-

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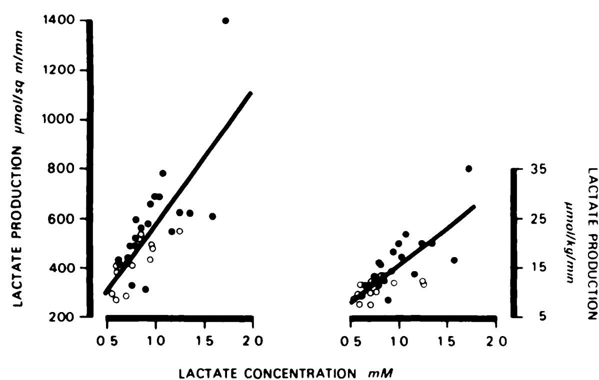

Chart 3. Relationship between plasma lactate concentrations and lactate production rates per sq m body surface area ($r = 0.77$, $p < 0.001$) or per kg body weight ($r = 0.78$, $p < 0.001$). ○, control subjects; ●, cancer patients.

pressed on the basis of body weight or body surface area (Chart 3). Also, the percentage of CO_2 derived from lactate oxidation and the rate of lactate oxidation were directly related to the lactate production rate (Chart 4). When control subjects were compared to the cancer patients, no significant difference was observed in the percentage of CO_2 derived from lactate oxidation (21.0 ± 1.6 *versus* 25.3 ± 1.7%). The percentage of the lactate production which was immediately oxidized was not related to the rate of lactate production, the mean values in the control subjects and cancer patients being 68.3 ± 2.7 and 62.1 ± 2.4%, respectively. Lactate disposal by mechanisms other than immediate oxidation, *i.e.*, the difference between the lactate production rate and the oxidation rate, was significantly increased ($p < 0.001$) in the cancer patients (227 ± 24 *versus* 128 ± 8 μmol/sq m/min or 6.0 ± 0.7 *versus* 3.2 ± 0.3 μmol/kg/min). The rate of nonoxidative lactate disposal was directly related to the lactate production rate (Chart 4). At least in part, conversion of lactate to glucose may account for the greater difference observed in the nonoxidative disposal in cancer patients in whom the percentage of glucose derived from lactate was significantly increased, (23.1 *versus* 15.8 ± 1.7%; $p < 0.01$). The percentage of glucose derived from lactate was also directly related to the lactate production rate (Chart 5).

No differences were found between patients and controls in free fatty acid, in fasting plasma glucose, or in immunoreactive insulin concentrations. Equally, values for respiratory quotient, O_2 consumption, and CO_2 production were similar (Table 1). Lactate production correlated poorly with tumor burden and no correlation was observed between increased lactate production and the following clinical or biochemical parameters: ambulatory status; degree of tumor differentiation; weight change; carcinoembryonic antigenic titer; hemoglobin; WBC; presence or absence of hepatic metastases; O_2 consumption; CO_2 production; respiratory quotient; free fatty acid; fasting glucose; or immunoreactive insulin concentrations.

DISCUSSION

Using an isotope tracer technique, we have shown that plasma lactate production and venous plasma lactate concentration are increased in patients with metastatic colorectal cancer compared to healthy, control subjects of comparable age and sex. With the exception of one patient, our values for lactate production are within the range (8.7 to 21.8 μmol/kg/min) reported by others for normal subjects and hospitalized patients with a variety of nonmalignant diseases (3, 11, 14, 18–20). To be noted is the fact that tracer techniques using [^{14}C]lactate measure only the production of nonlabeled lactate. Labeled lactate, derived from glucose or other compounds through recycling, would not be detected. Thus, lactate production rates reported in the literature and in this paper must be considered as minimal estimates of total production rates to the extent that recycling of [^{14}C]lactate occurs.

By using an appropriate group of controls, we have shown, by direct measurements, that lactate production is significantly increased in patients with metastatic colorectal cancer com-

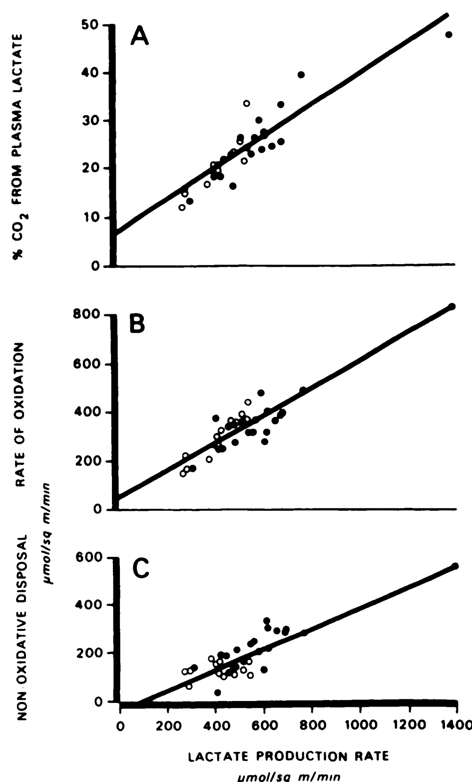

Chart 4. Correlation between the plasma lactate production rate and the percentage of CO_2 derived from lactate oxidation (*A*), the rate of lactate oxidation (*B*), and the rate of nonoxidative lactate disposal (*C*). ○, control subjects; ●, cancer patients. *A*, $r = 0.88$, $p < 0.001$; *B*, $r = 0.91$, $p < 0.001$; *C*, $r = 0.86$, $p < 0.001$.

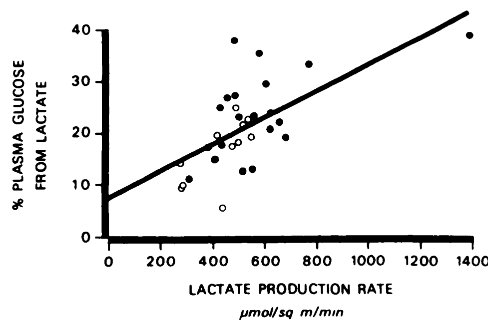

Chart 5. Correlation between the plasma lactate production rate and the percentage of plasma glucose derived from lactate. ○, control subjects; ●, cancer patients. $r = 0.63$, $p < 0.001$.

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Table 1

Hormone-substrate profiles and respiratory gas exchange

	Plasma glucose (mg/dl)	Plasma free fatty acids (μEq/ml)	Plasma immunoreactive insulin (μunit/ml)	O_2 consumption (mmol/sq m/hr)	CO_2 production (mmol/sq m/hr)	Respiratory quotient
Control group	91.8 ± 2.6[a]	0.892 ± 0.058	14 ± 4	324 ± 10	259 ± 9	0.80 ± 0.03
Cancer group	85.0 ± 4.3	0.889 ± 0.054	13 ± 4	311 ± 10	245 ± 9	0.79 ± 0.01

[a] Mean ± S.E. for 13 control subjects and 20 cancer patients.

pared to healthy adults. These data are in accord with circumstantial and direct evidence from the literature in which a predilection for increased rates of glycolysis by malignant cells or tissue has been described *in vitro* and *in vivo* (1). It is inferred but not proven that excess lactate production largely results from tumor glycolysis since increased anaerobic metabolism of glucose by human colorectal adenocarcinomas has been observed *in vitro* (15). Further, it has previously been shown that venous blood from tumor-containing organs generally has more lactate and less glucose than blood from comparable organs without tumor (2). This study does not exclude the possibility that remote humoral or nonspecific mechanisms are involved since direct arteriovenous lactate determinations across tumor tissue were not performed on our patients.

With the presumption that glycolysis is the principal source of lactate carbon, it follows that glucose metabolism may be altered in patients with increased lactate production. The highest recorded rate of lactate production in our study was 34.5 μmol/kg/min which is similar to a value reported by Kreisberg *et al.* (12) in a patient with tumor-associated hypoglycemia. This rate of lactate production could, if entirely derived from glucose, require 218 g glucose per day in addition to normal endogenous production for a 70-kg patient. Although our patient was normoglycemic, it can be appreciated from this analysis that, as suggested (12), hypoglycemia could occur in the absence of adequate continuing gluconeogenesis.

Oxidation represented an important mechanism for lactate disposal, accounting for at least 60% of the measured rates of lactate production in the control subjects and cancer patients. The moderately increased rate of lactate oxidation in the cancer patients testifies to efficient disposal mechanisms for this metabolite in the face of enhanced rates of production. At least 20% of expired CO_2 was derived from the immediate oxidation of plasma lactate in both groups of subjects. These high values are probably due to conversion of lactate to glucose which is oxidized and to equilibration of infused [^{14}C]lactate with the pyruvate pool through which a rapid flow of carbon from major energy-yielding fuels could be anticipated.

Rates of lactate disposal by mechanisms other than immediate oxidation were increased 2-fold in the cancer patients compared to control subjects. We and others have previously shown that Cori cycle activity, in which glucose is metabolized to lactate which is resynthesized to glucose, is elevated in some cancer patients (7, 8, 22, 23). The finding in this study of a significantly increased percentage of glucose derived from lactate in cancer patients compared to controls is consistent with, although mathematically distinct from, these observations. The resynthesis of glucose from lactate may represent an important disposal pathway for lactate which, under circumstances of high glucose consumption by tumor, may be essential to preserve euglycemia.

Although high rates of glucose utilization are considered to be characteristic of the neoplastic cell, we have previously shown that endogenous glucose production in cancer patients is only moderately increased (7, 8). In this regard, Waterhouse (22) has commented that even normal glucose production seems inappropriately high in patients with anorexia, weight loss, or reduced caloric intake due to tumor. Glucose turnover rates were not, unfortunately, measured in this study but are likely, on the basis of the patient population studied, to be similar to our previous results (7, 8). It seems probable that increased lactate production from glucose accounts for the moderate increase in glucose turnover previously noted and that, without increased gluconeogenesis, endogenous glucose production might be normal or low.

The apparently poor correlation between excess lactate production and tumor burden was surprising. It is noteworthy that normal lactate metabolism was observed in some patients with bulky tumors while the reverse was occasionally true of fully ambulatory patients with minimal detectable tumor. This observation seems consistent with the known exceptions to Warburg's generalization that glycolysis is greater in neoplastic than in normal tissue (21) and suggests, directly or indirectly, different degrees of metabolic differentiation. This study emphasizes that metabolic heterogeneity with respect to lactate metabolism exists within an apparently homogeneous group of patients with colorectal cancer.

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