THE CHEMICAL PROTOTYPE OF TUMORS IN RELATION TO THE COMPOSITIONS OF RAT TISSUES

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Introduction

Investigations have recently begun to demonstrate some order among neoplasms, at least among those experimental tumors of the rat that are most often studied by biochemists. Before this, biochemistry contributed to cancer research a succession of fragile hypotheses -- D-glutamate in tumor proteins, defective tumor respiration, or unique constituents of tumors -- which after enormous effort were one by one proved to be illusory. Failures rather than knowledge accumulated. In consequence, neoplasms are still considered by many to be chemically bizarre and disordered growths, even in the face of evidence that they do grow and reproduce only too efficiently. Biochemical "successes" in cancer research will remain essentially negative in character until the orderliness, and not the disorder, of tumors is studied.

A modest beginning is the evidence to be presented here that a limited spectrum of tumors share a common composition, akin to that of normal fetal tissues. The quantitative chemical similarities between tumors and immature tissues fit their functional likenesses. The qualitative similarity of many specific isozymes in tumors and immature tissues also shows that there is reactivation in tumors of a significant portion of the cell's fetal genome. Constructive hypotheses instead of only null hypotheses about the nature of neoplasms can now be tested.

Histological Characterization of Neoplasia

Morphological identification and classification is the rock upon which our knowledge of neoplasia rests, inadequate though that may seem to some.

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Constructive chemical ideas about the nature of neoplasms can reasonably be expected to accord with what is already known of the morphological characteristics of tumors. Chemical information that cannot be related to the morphological experience of a hundred years may indeed be irrelevant to the nature of neoplasia. Initially, at least, chemical information must be used in a special way to meet this elementary criterion of relevance to the only coherent knowledge of tumors we now have.

Morphology teaches us that neoplasms are altered cells of the host -- flesh of our flesh -- which can (usually) be classified as coming from one or another of the normal tissues of the body. Those coming from any single tissue are found to grade from nearly normal, well-differentiated tumors up to such undifferentiated tumors that classification as to origin is not possible. In general, "malignancy" or neoplasticity increases with the higher grades of tumor dedifferentiation and faster rates of growth. The spectrum of graded tumors from any one tissue type and the many types of normal tissues that can become neoplastic produces a very great variety of neoplasms. Each tumor may in fact be unique, and if so, the efficiency of the histological classification system is all the more impressive for containing in a practical way an almost infinite variety of tumors.

Can biochemical, as opposed to histological characterization, confirm or even extend this successful scheme for recognizing and classifying tumors? Quantitative chemical analysis is a powerful tool of very high resolution. It can demonstrate numerical relations or differences between tissues. The kinds of analyses used can be multiplied to whatever extent is necessary to reveal even minor distinctions. It is necessary only to express the different

components on the same scale so that they can be compared. The averaged chemical differences of a number of components measured in any two tissues can be expressed as a number, and this can be graphed as the "chemical distance" between the tissues. A map is produced of the relationships among tissues. If the histological characteristics upon which microscopists rely could be quantified, the "histological distances" between tissues and tumors could also be graphed in an exactly analogous manner. This would show the structure of the classification system already described. In fact, no such numbers exist, but a diagram of the classification will show its principle, and also show that it has unresolved ambiguities. Pathologists divide fairly equally between favoring one or another of the two possible conceptual structures shown in Fig. 1. The difference between the structures is not trivial. It would be important for biochemistry merely to confirm the general nature of these conceptual structures derived from histology. It would be a significant scientific advance to identify which one is correct.

I have assumed in these conceptual structures that there is a graded relationship between different tumors of a single tissue type, because such gradations are supported by strong histological and clinical evidence for a number of important kinds of human tumors, and because chemical grading of the rat hepatomas has been a major result of recent biochemical endeavor. Gradation within a single tumor type is shown diagrammatically in the figure by the linear, monotonic departure of tumors (open circles) farther and farther from their tissue of origin (solid circle). The differences ("distances") between tumor and parent tissue become greater for the higher grade tumors. Both conceptual structures also show distances among the

several tissues of origin, reflecting the familiar differences, histological and chemical, that exist among liver, kidney, skin and so on.

The crucial difference between the two conceptual structures in Fig. 1 is that the tissue-tumor relations in the "unique" structure are peculiar for each tissue (shown by the tumors lying in different directions from their tissues of origin). In the second "concerted" structure all tumors depart in the same general direction from the several normal tissues. If the first structure were correct, neoplasia of each tissue would constitute an individual problem, with its own nature and means of diagnosis and treatment, and little transfer of information between problems could occur. The second structure offers a more general set of solutions that would be applicable to tumors of many kinds of tissues. The success of a pathologist in recognizing many kinds of tumors, including ones from tissues not previously seen by him, suggests that some general solutions do exist.

It is important to note a variant of the second conceptual structure (indicated by dashed lines). Here tumors depart in nearly the same direction from the normal tissues, but in such a way that the higher grade tumors actually become more like one another. This subsidiary scheme could explain why even the most experienced pathologists sometimes cannot identify the cellular origin of a particular tumor. The highest grade tumors (labelled "prototypic") will have lost most of the characteristics of normal tissues and thereby simply will have become indistinguishable. Such chemical similarity between tumors from different sources (all probably of high grade) was in fact demonstrated by J. P. Greenstein a quarter of a century ago (1; Summary and references in 2, Chaps. I and II). Fig. 2 shows Greenstein's

summary (3) of the chemical similarities among different mouse tumors and the contrasting chemical diversities found among normal tissues. This uniformity of the chemical compositions of different kinds of tumors contradicted the general appreciation that there was extreme biological diversity to be found among neoplasms. One solution to this still unresolved paradox of uniformity versus diversity among tumors is inherent in the subsidiary second diagram of Fig. 1. Uniformity exists among the high grade tumors. Diversity exists among the lower grade tumors. There is dissimilarity between two tumors that are each closely related to a different normal tissue, and also among those tumors of different grades from a single tissue.

This brief review of the one coherent body of information about the nature of neoplasms, that of morphology, shows that important alternatives in its conceptual scheme remain. Biochemical information is compatible with that of morphology in the single, well-investigated case of the different grades of hepatomas. During the past decade the orderly chemical gradation found among tumors of this single organ has been firmly established under the leadership of Weber (4, 5). Any single line in any version of the schemes in Fig. 1 can represent this relationship among liver and the hepatomas. For this single tissue-tumor system biochemistry is in accord with histology. This is an important achievement for biochemistry, even if it is not an advance for oncology.

The chemical uniformity among different kinds of tumors as reported by Greenstein remains paradoxical, but it could be fitted into one of the alternatives still open in the morphological scheme. Specifically, an orderly chemical grading in another tissue-tumor system besides that of liver, if

found, would also show, by comparing the two systems, whether the two kinds of high grade tumors were more alike than their parent tissues. This possibility is represented by any two lines in the subsidiary second version (dashed lines) in Fig. 1.

Chemical Characterization of Neoplasia

It is possible to construct experimentally a map of the relations among tissues such as that diagrammed in Fig. 1. Average chemical differences between two tissues represent distance on the map. Examples of this have been published (2, 6) and the mathematical techniques are widely used in other fields (7,8). The main needs are analyses of a sufficient number of components in every tissue to be compared. Systematic analyses of even single tissues are almost unknown in biochemistry, because its work has been partitioned among different tissues, ages, sexes and species of animals. Obviously, the quantitative analyses must be reliable and reproducible. Because enzymes characterize uniquely both the composition and function of tissues, they are the components of choice for such comparisons. There is more confusion than there ought to be in the minds of some biochemists about the validity of assays of enzymes in tissues by catalytic activity under standardized conditions. However, measurements of relative enzyme concentrations in different tissues can be reproduced in any laboratory using any adequate assay of that enzyme (9, 10). The results are therefore accumulatable, and this will lessen the cost and increase the range of subsequent studies.

A careful selection of tissues and tumors is necessary to show the significant relationships with an utmost economy of analyses, because even

the minimum number of analyses is a prodigious task. Transplanted tumors of the liver and mammary gland were chosen that represented in each case a spectrum of grades according to morphology and growth rate. Three different states of the mammary gland itself were analyzed because of uncertainty about which (virgin, pregnant and lactating glands) should be considered the tissue of origin of tumors. The results, surprisingly enough, turned out to be almost unaffected by the state of the gland. A number of other normal, adult and fetal tissues were subsequently analyzed to provide a background against which to view the tissues and tumors of primary interest.

A simpler preliminary study was encouraging (6). A random selection of enzymes with robust quantitative assays were measured. The amounts of 21 enzyme components in the critical tissues and tumors showed the chemical relationships among the tissues of the liver-hepatoma series and the mammary gland-mammary carcinoma series to be as indicated by dashed lines in Fig. 1. That is, undifferentiated tumors from the two organs were very similar; differentiated tumors were less similar, and the two parent tissues were unlike (6).

We have since increased to 30 the number of components analyzed, included other normal tissues in the comparison, checked many of the analyses, and refined the numerical analysis (by the use of the logarithmic distribution among tissues of the values for a component and by the use of principal component analysis (11)). These were conservative modifications that provided a rigorous and objective test of the similarities initially found. The main differences between tissues in the latest study are graphed as distances in Fig. 3. The agreement with the subsidiary conceptual structure shown by

dashed lines in Fig. 1 is close enough so that other alternatives for these two kinds of tumors can now be abandoned. The gradation, from left to right, of liver, slow- and fast-growing hepatomas confirms the monotonic relation within this series that has been repeatedly established by studies of one or a few components at a time to a total of well over a hundred substances (2). Some components are invariant in this series, but others either decrease or increase monotonically from liver to undifferentiated hepatomas (4). There is a similar gradation from left to right of mammary gland, differentiated and undifferentiated mammary carcinomas. It is not limited to hepatic tissues that chemical compositions of tumors change in parallel with loss of differentiation and increase of growth rate. Furthermore, the direction of gradation in both series means that the same general kinds of chemical changes occur in the different tumors. Finally, the two tissues of origin are chemically very different, but the tumors are less so. The highest grade tumors of the two kinds are very alike.

Relationships between tissues shown in Fig. 3 are remarkably stable. Half as many components give the same relationships and almost the same distances. The vertical and horizontal dimensions in Fig. 3 represent the two major ways in which two dozen rat tissues and tumors differ from one another. Each dimension encompasses a combination of analytical chemical values, and the components affecting one dimension more than the other can be ascertained. The vertical dimension distinguishes between normal adult tissues. Other such tissues besides liver and mammary gland are widely distributed in the vertical dimension, and are also to the left of the figure. Other kinds of tumors are more or less narrowly focused to the right of the

figure. Fetal tissues are also narrowly distributed to the right of Fig. 3 near the undifferentiated tumors. Fetal lung is shown as representative of the cluster of fetal tissues that surround it. Immature tissues (5 days after birth) are midway to the adult tissues. The fetal tissues are closer together and therefore chemically resemble each other more than do the adult tissues into which they differentiate, as would be expected. They are near (i.e., similar) to the undifferentiated tumors, which also resemble each other more than do the tissues from which they dedifferentiated. Clearly, the horizontal dimension of Fig. 3 is an axis of differentiation-undifferentiation. The most important components affecting location on this axis can also be identified. It is apparent that these chemical analyses show meaningful interrelationships among tissues, and that selected analyses, such as those locating tumors toward the right, could help distinguish between normal and cancerous tissues.

There can be no attempt to balance the chemical characterizations of these two tissue-tumor series in the rat with the accumulated histological characterizations of all tumors that now fill huge libraries. It is more to the point that observations in another discipline, such as chemistry, has quite readily confirmed the orderliness, based on tissue cell type, that histologists created among the welter of tumors they have examined. There are gradations between a normal tissue and its most undifferentiated tumors that are confirmed by these independent observations. So chemical measurements join morphological signs of dedifferentiation and anaplasia and the differences in behavior such as growth rate as indicators of the greater or lesser neoplastic character of a tumor. That there are such gradations is

recognized by calling tumors "benign" or malignant". The fact can be stated more objectively and simply: there are various degrees of neoplasticity among tumors of any given kind. This is established histologically for at least some kinds of tumors, and it is well-established biochemically for rat hepatomas. Perhaps now this scaled nature of tumors should be recognized as a central and not a peripheral characteristic of the phenomenon of neoplasia.

In addition, assuming that what has been shown for these two kinds of experimental rat tumors is generally so, the new chemical observations add to the resolution of tumors from tissues in important ways. These are the relations between tumors of different kinds and between tumors and immature tissues as shown in Fig. 3. Despite the chemical differences between various normal tissues, tumors of these tissues differ from the parent tissues in much the same chemical ways (i.e., in the same direction in Fig. 3). Although prescient pathologists have been aware of this possibility, objective evidence for the "concerted" and against the "unique" structures of a tumor classification was lacking. Fig. 3 now points to an important unity among neoplasms of all kinds. The unity shared by different kinds of neoplasms consists of departures in the same direction from normal and ending in a common type of tumor. The most undifferentiated tumors from both tissues are so very similar that the existence of an "ideal" or prototype of tumor composition is suggested. At the same time, the old paradox of similarity and diversity among tumors is resolved by the varied degrees of differentiation of the tumors. More differentiated tumors can be looked upon as greater or lesser dilutions of the prototypic tumor with the composition of a particular normal tissue. Such admixture results in closer similarity of a tumor

to its own parent tissue, greater chemical diversity between different kinds of tumors, as well as moderation of a tumor to one that is less neoplastic in its behavior, form and composition.

The relationships between adult tissues and tumors that were derived from studies of which Fig. 3 is only a part can be represented diagrammatically as the surface of a cone (Fig. 4). From the center of its base (more accurately it is an indented base) the fetal tissues differentiate and diverge to become the separate adult tissues along the rim or conic perimeter. Tumors lie along the conic surface with the least dedifferentiated ones close to the rim and the most dedifferentiated ones at the apex. The latter are on the axis with fetal tissues and not very far (not very different) from them, since the indented base of this structure is more accurately a cone within the larger cone. Embryonic characteristics of tumors have often been noted, and as often denied to be "truly" embryonic in character. The objective chemical similarity of prototypic tumors to embryonic tissues is, however, not unexpected. Although fetal liver is more differentiated and less like prototypic tumors (2) than most fetal rat tissues, Greenstein saw long ago the chemical similarities of hepatomas to fetal liver (1). Correlational analysis of his data confirmed his conclusions (2: Chap. II). The relation to other fetal tissues is closer. In merely logical terms, when the specific chemical compositions that distinguish tissues are lost by tumors and they must move from the rim of the cone in Fig. 4, these dedifferentiated tumors could only come to resemble the still undifferentiated embryonic tissues.

Qualitative Chemical Similarities of Fetal and Tumor Tissues

Similarities between the quantitative patterns of components form the basis of the relationships between adult and fetal tissues and tumors shown in Figs. 3 and 4. Such a relation first emerged from intensive study of the liver-hepatoma series. Now it has been generalized to the mammary gland-mammary carcinoma series. The same sequence of hepatic and then other studies led to recognition of qualitative chemical characteristics which can specify the nature of tissues concretely. Observations on the occurrence of isozymes in hepatic tissues, often made in different laboratories, indicated that there was a rule governing the type of isozyme found in fetal and adult livers and in hepatomas (2: Chap. VIII). If fetal and adult rat livers had different forms of an enzyme, the fetal form was the one found in hepatomas. The only exceptions were rare and single occurrences of an adult isozyme in the most highly differentiated hepatomas. Some isozymes, like the M form of lactate dehydrogenase, occur in both fetal and adult livers, and in these cases, consistent with the rule, the same type of isozyme was also found in hepatomas. Subsequent work that was aimed specifically at testing this rule for liver and hepatomas has furnished us with additional examples (12). Following the sequence of study of hepatomas first, then of other tumors, it was important to know if the same rule held beyond the hepatomas. Could isozymes be identified in other immature tissues and were these replaced in the adult tissues, only to reappear in tumors of those same tissues?

Electrophoretic analyses of three isozyme systems were made in a number of rat tissues (Fig. 5). A particular form of each enzyme could indeed be recognized as the one predominating in all of the various fetal tissues examined. The same isozymic forms also predominated in tumors derived from

liver, kidney, muscle and mammary gland (13). The forms of the three enzymes common to fetal and tumor tissues are listed in the lower half of Fig. 5. A different form of one or two of these enzymes predominated in each of the normal, adult tissues. These are enclosed in boxes in the upper half of Fig. 5.

The results illustrate the same rule that holds for hepatomas, that these other immature and neoplastic tissues also share the same isozymes while the cognate adult tissues have at least some different forms. Investigation of still other enzyme systems should reveal more qualitative chemical evidences of differentiation in normal, growing tissues. Such isozymes would, perhaps, also be lost in tumors, and more such examples would increase the precision with which a tumor could be qualitatively distinguished. With only three enzymes, adult tissues showing rapid cell growth such as regenerating liver, small intestine and spleen had isozyme patterns only marginally different from that of fetal and tumor tissues.

Thus, like the quantitative comparisons, the qualitative chemical patterns of isozymes also show similarities between fetal and tumor tissues, and differences of both from adult tissues. The nature of the isozymes as specific proteins under genetic control allows a further conclusion about the tissue relationships. Certain parts of the genome that are active in mature tissues are turned off in tumors. Another part is activated. That part activated in tumors is normally active during tissue immaturity. In both immature and tumor tissues this part of the genome makes the same recognizable isozymes.

Chemical Distinctions of Tumors

A very large number of tissue components have had to be measured and identified in each of a number of fetal, adult and tumorous tissues to see the chemical relationships between these tissues. Still more assays will be needed to extend the findings to other tissue-tumor systems and to reveal, amongst the similarities, what particularly distinguishes tumors from normal tissues. Among the randomly chosen components needed in number to characterize each tissue, some, like those that move tumors to the right of adult tissues in Fig. 3, are of greater interest than others. Such components can be identified by the weights (factor scores) they have in each dimension of the factor analysis (11) from which Fig. 3 is derived. Specially interesting components can also be identified independently by stepwise discriminant analysis (11). This is a statistical technique that seeks to classify the tissues correctly into groups using the fewest possible components. The results can be graphed to show the degree of separation between group means and the dispersion of the groups. Seventeen normal (adult and fetal) tissues and seven tumors, among which those in Fig. 3 are included, can be separated in a significant manner by using only four of the thirty components that were measured. The separation of the two groups and the dispersion of the individual tissues within each group are shown in Fig. 6. The four enzymes which allow this clear distinction between normal and tumorous tissues happen to be the most efficient for this purpose among the thirty measured components. They are not otherwise remarkable, and doubtlessly still other components can be found that are as good or better discriminants, especially if particular isozymes are examined for this task. Fig. 4 does demonstrate, however, that there are usable chemical differences between sizable groups of

normal and neoplastic tissues as was implied by the structures of Fig. 3. The separation is all the more remarkable because of the similarity of fetal to tumor tissues. It was therefore of interest to attempt to distinguish all three groups of adult, fetal and tumor tissues. This required only five enzyme measurements for a significant separation. It is shown in Fig. 7. Some of the enzymes used are common to Figs. 6 and 7. In general, relative abundance in certain tissues of each component may have a complex role of tightening one group or separating two groups. The method offers a way to screen out certain components for more detailed study of their contributions to the different natures of normal and neoplastic tissues.

Discussion

A new genre of biochemical investigations was necessary to gain the perspective shown here of the relations between rat tissues. Qualitative analyses have previously sufficed to construct metabolic maps showing the chemical unity of living systems. Quantitative comparisons of the catalytic activity of an enzyme within a laboratory have allowed many of them to be purified, and have permitted the regulation of their relative concentrations in a tissue to be studied. Now qualitative identifications and relative quantifications of enzymes in each of a series of tissues show the large variations on the theme of chemical unity that can be found among tissues that function differently. The individual enzymic patterns reflect these varied functions because of an intimate relation that exists between an enzyme and a cell's activity. Enzyme analyses are measurements of the things by which cells differ. The analyses are meaningful because they can be reproduced in other laboratories and fitted to cell functions. The results

can be accumulated, corroborated and their implications reexamined. Systematic analyses of many tissues are needed in addition to the few rat tissues described here. This can only be accomplished by cooperative work in many laboratories. The preliminary results indicate that the effort will be worthwhile.

Cooperative biochemical work related to cancer in many laboratories over the past decade has focused on the rat hepatomas, and specifically those graded in neoplasticity between the extremes of adult liver itself and undifferentiated, fast-growing hepatomas. Given Greenstein's summary that all tumors were similar, it was essential that the chemical variety to be found among tumors of a single tissue be demonstrated. The huge success of this inquiry may be overlooked by failure in the ostensible goals of that cooperative effort (14): to detect some minimal deviation among the normal variations of liver that would epitomize cancerousness, and to do so by ignoring the full-blown deviations that are plain and obvious in the highest grade tumors. Preoccupation with minimal changes from the normal crumbles as a strategy when a second kind of normal tissue is investigated. What tumors of two tissues do have in common are those characteristics shared by tumors regardless of origin and that appear most recognizably in the "progressed", undifferentiated prototypic tumors. Disinterest in the most "progressed" and altered tumors has caused them to be overlooked as the reference standard for neoplasticity in its purest form. This is what the prototypic tumor appears to be. Differentiated tumors appear to be diluted versions of that prototype -- diluted by the composition of one or another normal tissue in the tumor series corresponding to that respective tissue.

The interpretations of these biochemical results draw notable support from their parallel with the general histological description of neoplasms. Both approaches are immeasurably strengthened by this concordance. The biochemical results have also extended or made more precise some classical histological views by demonstrating the similarity of undifferentiated tumors from two sources, by recognizing the histological grades as steps in the series culminating in a prototypic kind of tumor, and by furnishing objective evidence for the long suspected relation between tumors and normal, immature tissues.

This biochemical description of tumors is based upon their enzymic elements. Enzymes are intimately connected in one direction to the function of the cell, and in the other direction to the qualitative and quantitative expressions of the cell's genome. In these two directions, toward the biological nature of tumors and toward the genesis and regulation of them, are central areas of our ignorance of the cancer problem. Beyond the task of deepening and extending the chemical relationships between tissues and tumors that has been outlined here, we must address new formulations of old problems lying in both directions. Looking toward the biological nature of tumors, it would be helpful to design diagnostic and therapeutic modalities with the characteristics of the prototypic tumor in mind. In the other direction, we must learn what keeps one specific part of the genome active in an adult tissue, so that its state of differentiation is maintained. Future studies, necessarily including immature tissues as controls, should reveal the salient differences between the normal and abnormal undifferentiated tissues.

Summary

Analyses of the characteristic constituents (e.g., the relative amounts of enzymic components) of rat tissues and tumors have been used to measure the degrees of chemical similarity among the tissues. Undifferentiated, fast-growing tumors originating from the liver and from mammary gland have almost the same compositions. Differentiated tumors from the same sources are more unlike in compositions, each kind tending to resemble the chemically very different parent tissues. A prototypic composition of tumors, whatever the source, appears to exist. When this is diluted by the various compositions of normal tissues, the products are tumors of lesser neoplastic character (more differentiated, slower growing, less autonomous tumors).

The prototypic composition of tumors is very similar, but not identical to that of many fetal tissues in both the quantitative patterns of enzymes and in the qualitative identities of certain isozymes. It appears that a fraction of the cell's genome acts in the same way in immature and tumorous tissues, but differently in adult tissues. Measurement of selected enzymes that are part, or are not part, of the prototypic composition of tumors can distinguish a variety of tumors from a variety of normal tissues. Relatively small numbers of enzymes (4 or 5) are sufficient to make this important distinction between the 24 normal and tumorous tissues examined.

A certain orderliness is now evident among the natures of tumors studied. Methods like those used here can focus attention upon the chemical components and processes that contribute most relevantly to the nature of neoplasms.

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Captions

Fig. 1. Alternative conceptual schemes of the relations between neoplasms and their tissues of origin, based on the accepted histological classification system. Average differences between tissues and tumors show as distances on the map, in directions related to the kinds of differences. Individual normal tissues (0) are connected through increasing grades of their neoplasms to the most undifferentiated tumors (0). In the left-hand, "unique" model, the tumors each bear unique directional relations to their normal tissue of origin. In the right-hand, "concerted" model, all tumors differ in the same direction from normal tissues. A specialized version of the latter scheme (---) shows no distinctions remaining between the most undifferentiated tumors from all sources ("prototypic tumor").

Fig. 2. Relative enzyme activities in normal and neoplastic tissues of mice. Lengths of horizontal bars in a column represent levels of a given enzyme. (From Greenstein (3)).

Fig. 3. The kinds and amounts of the main chemical differences between normal rat tissues (0) and tumors (0). The two dimensions (small arrows indicate the less important third dimension) from a principal component analysis of 30 components measured in twenty-four tissues account for nearly half of the variations between all tissues. Mammary glands (VM, virgin; PM, pregnancy; L3, third day of lactation) and mammary carcinomas (1C, 3230AC, 5A and Walker in order of increased dedifferentiation) are above the hepatic series of liver, slow-growing Morris hepatoma 7800, and fast-growing hepatomas (F. Hep.). Fetal lung (Fe Lung) is representative of most fetal tissues.

Fig. 4. Geometric projection illustrating the relationships between tissues, with their average chemical differences expressed as distances. Fetal tissues (FT) arise from the primordium at the hub of the wheel and diverge along the spokes into the differentiated adult tissues at the rim. A single tissue gives rise to a variety of tumors which can be arranged in order of loss of differentiation along the surface of the cone, and tumors from various tissues form series that converge toward the apex. The axis of the cone defines the most undifferentiated state of both normal and neoplastic tissues. (From Knox (6)).

Fig. 5. Predominant isozymic forms of three enzymes found by electrophoresis in fetal, adult and tumor stages of four tissue systems. Each isozyme is designated by a familiar letter. The identical patterns in fetal and tumor tissues are superimposed in the lower half of the table (no fetal version of mammary gland was examined). Boxes enclose the isozymes distinguishing differentiated (upperhalf) from undifferentiated states of a tissue. (From Farron et al. (13)).

Fig. 6. Statistically significant distinctions between groups of normal and neoplastic rat tissues made by discrimination analysis (11). Tissue contents of the listed enzymes provided these separations (F-value is given) between the group of normal fetal and adult tissues (indicated by numbers at right) and the group of mammary carcinomas and hepatomas (labels at left), each about a group mean (*).

Fig. 7. Statistically significant distinctions of the three groups of fetal, adult and tumor tissues. Same analysis as Fig. 6. The fetal tissues (1-4) are liver, kidney, brain and lung, respectively.

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Unique Model

2 Concerted Models

Kinds of Differences between Tissues (●) and Tumors (○)

4 kg 2

Fig. 3

Fig. 4

PATTERNS OF THE PREDOMINANT ISOENZYMES
IN ADULT, FETAL AND NEOPLASTIC RAT TISSUES

	Liver	Kidney	Muscle	Mammary Gland
<u>Adult Tissues</u>				
LDH	M	H	M	M(H)
Aldolase	B	A(B)	A	A(C)
Pyruvate kinase	L	K	M	K
<u>Fetal and Tumor Tissues</u>				
LDH	M	M	M	M
Aldolase	A	A	A	A
Pyruvate kinase	K	K	K	K

Fig. 6

Fig. 7