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Do we need to redefine a cancer metastasis and staging definitions?

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Abstract

Metastasis is the most lethal attribute of cancer cells and clinical decisions regarding treatment are based largely upon the likelihood of developing metastases. However, improvements in detection as well as recent experimental data have raised questions about the most appropriate definition of a metastasis, especially whether the mere presence of cells at secondary sites constitute a metastatic lesion. After reviewing the experimental basis of metastasis, a definition of metastasis is proffered along with a proposal to consider regarding modification of staging parameters.

When cancer survivor Clifton Leaf lamented the state of cancer research in early 2004, he concluded that progress in the “War on Cancer” was limited because research into the prevention and treatment of metastasis had been inadequate (1). He asserted that failure to control establishment of secondary colonies was the major contributor to failure and that winning the war against cancer would require increasing resources devoted to studying metastasis.

What is a metastasis?

At first, such a question would seem elementary. Yet, the answer is not as straightforward as one would first think due to improved detection methods and new experimental findings. Both categories of new data necessitate re-evaluation of long-held paradigms that, in turn, initiate a domino effect on both clinical cancer control and the design and interpretation of experiments studying metastasis.

What has prompted me to re-examine the definition of a metastasis?

Perhaps a new definition would not be needed were consensus about the properties of a metastasis self-apparent. Ultimately, there is honest disagreement among researchers, oncologists and pathologists about what constitutes a metastasis. One of the reasons metastasis is so difficult to define is that the same word describes the process and the outcome. This chapter focuses on the definition of ***a*** metastasis (i.e., the product), rather than defining the process of cancer spread itself, although the two are inextricably linked.

The primary impetus for revisiting the definition of a metastasis is recent experimental data arising from characterization of metastasis suppressors. In many cases, cells expressing metastasis suppressors disseminated with equal efficiency to the corresponding parental cells; however, they failed to colonize secondary sites. The data prompted the question: What

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constitutes a metastatic mass? Given that it is now increasingly possible to detect single cells at secondary sites, should staging criteria be modified to accommodate new methodology?

In order to build a definition of a metastasis, it is first essential to disavow misconceptions regarding antecedent steps and the outcomes. Five myths regarding metastasis have crept into the scientific and medical literature because of imprecise use of terminology.

Myth #1: Metastasis is an inherent property of cancer cells—In a 2000 review, Hanahan and Weinberg describe six hallmarks of cancer cells (2). Besides, immortality, abnormal growth regulation, self-sufficient growth, evasion of apoptosis and sustained angiogenesis, invasion and metastasis were listed as distinguishing characteristics. Unfortunately, some misinterpreted the list as meaning that all tumors are invasive and/or metastatic, which is certainly not the case.

Myth #2: Metastasis and invasion are equivalent phenotypes—Some tumors are highly aggressive, forming secondary lesions with high frequency (e.g., small cell carcinoma of the lung, melanoma, pancreatic carcinoma); whereas, others are rarely metastatic despite being locally invasive (e.g., basal cell carcinomas of the skin, glioblastoma multiforme). Therefore, metastasis is not an inherent property of all neoplastic cells or even invasive cells. Likewise, while invasion through a basement membrane is the hallmark which objectively defines malignancy, not all neoplasms are invasive (e.g., carcinoma *in situ* of the breast (DCIS), prostatic intraepithelial neoplasia (PIN)). Some might argue that morphologic criteria are objective measures of malignancy; however, the discordance between experienced pathologists for grading (and even diagnosis) can be great (e.g., (3-5)), suggesting that those parameters might be more subjective in nature. In short, not all tumors are invasive and metastatic and less subjective criteria are needed.

Myth #3: Metastases arise only from cells disseminated via the blood or lymphatics—Most commonly, metastasis is described in terms of hematogenous dissemination. However, secondary tumors can arise because tumor cells have migrated via lymphatics (i.e., lymph node metastases are extremely common in many carcinomas) or across body cavities (e.g., ovarian carcinoma cells most frequently establish secondary tumors by dissemination in the peritoneum while rarely forming metastases via hematogenous spread). Lugassy and colleagues recently documented dissemination of melanoma cells along the space between endothelium and basement membrane (6) (i.e., the cells do not appear to enter the vascular lumen *per se*). This route of dissemination is reminiscent of perineural spread common in pancreatic and prostatic carcinomas. Thus, the route of dissemination is not inherent to a definition of metastasis.

Myth #4: Tumor cells at secondary sites are metastases—Even cells that have been selected for metastatic capacity exhibit low efficiency for developing metastasis, seldom exceeding 0.01% (7,8). Entry of cells into the blood stream is apparently not uncommon. In fact, more than a million cells per gram of tumor can be shed daily (9). Fortunately, establishment of secondary foci is much less frequent. This point was elegantly demonstrated by Tarin and colleagues using peritovenous shunts to palliate ascites burden for women with ovarian cancer (10). Although millions of tumor cells were directly deposited into the vena cava daily, the women did not develop secondary blood-borne tumors with higher frequency.

Myth #5: Extravasated cells are metastases—The fate of blood-borne tumor cells is somewhat controversial because of apparently contradictory experimental evidence. Fidler showed, using radiolabeled tumor cells, that the vast majority did not survive in the circulation (8,11). Several other labs subsequently demonstrated that cell killing occurred because of sheer (12,13) or immune selection (14,15). Recently, however, Chambers, Groom, and colleagues

have quantified tumor cell fate using an intravital microscopy accounting method (16,17). In contrast to the above data, they found that the majority of cells not only survived, but extravasated. Explanations for the disparity include different cell monitoring methods (i.e., radiolabeling *vs* fluorescent tagging, respectively). The latter explanation is not satisfying, however, because we observed similarly rapid clearance of tumor cells using green fluorescent protein-(GFP) tagged cells (18), indicating that the explanations are not so simple.

Muschel and colleagues used intravital microscopy to demonstrate that, in their model, the majority of cells not only remained intravascular, but began to proliferate intralumenally (19). The intravital microscopy data, while in apparent conflict, may not be contradictory. The experiments measured colonization in two different organs (liver and lung) and used different tumor cell lines (mammary carcinoma and melanoma), emphasizing heterogeneity for both tumor and host parameters. The findings are most easily reconciled by the explanation that mechanisms of metastasis will be tumor type-dependent, colonized tissue-dependent and host status-dependent. In other words, there are multiple mechanisms and support pathways for cells to colonize secondary sites. As with most tumor cell properties, there will be a spectrum of behaviors.

Unfortunately, insufficient data exist to quantify the fraction of tumor cells successfully seeding secondary tissues once they have entered the vasculature, especially in human cancers. Regardless, all studies show that the majority of cells entering the vasculature fail to form macroscopic foci. Furthermore, of the cells arresting in tissues, the majority fail to colonize non-native tissues.

Hallmarks of metastatic cells

Springboarding from the hallmarks posited by Hanahan and Weinberg, the following are submitted as hallmarks of metastatic cells. Such characteristics are found only within a subset of cells already exhibiting the posited hallmarks of tumor cells (2).

Invasion: Neoplastic cells must be capable of migrating away from the primary tumor. To accomplish this, they utilize a variety of proteinases (e.g., MMPs, cathepsins, uPA, etc.) and motility mechanisms (20-22). Similar molecules are involved in extravasation once the cells have transited elsewhere. However, invasion need not require production of proteinases as has been commonly believed. Friedl and colleagues recently showed that tumor cell migration through collagen matrices using amoeboid motility mechanisms still occurs in the presence of proteinase inhibitor cocktails, thought to inhibit most proteinases (23). Their results highlight how long-held notions that invasion requires proteinases must now be questioned. They further illustrate how the outcome - a metastasis - can be separated from the process of dissemination. In addition, the intuitively obvious assumption that most proteinases within tumors are tumor cell-derived has been challenged by immunohistochemistry and *in situ* hybridization studies showing that most proteinases are produced by stromal cells (21,24,25).

Dysregulated adhesion: Tumor cells must detach from the primary mass and then re-adhere at a locale(s) discontinuous from the primary tumor. Using hematogenous metastasis as an example, it is important to recognize that the transit time from the tips of the toes to the brain is seconds to minutes, meaning that the cells could not shut down expression of all adhesion molecules as they depart the primary tumor and re-express them using conventional transcription/translation exclusively when they arrive at secondary sites. Cells may use alternative adhesion molecules or may selectively alter adhesion by post-translational modification of already-expressed

proteins, glycoproteins, lectins or other molecules. Perhaps, they could even alter intracellular signaling from surface adhesion molecules.

Survival: Dissemination, as implied above, requires tumor cells to detach from whatever matrix or cell-cell anchor(s) are involved in tissue structure. Under normal circumstances, epithelial cells undergo apoptosis/anoikis when adhesion is limited (26). Therefore, metastatic cells must be, at least relatively, resistant to anoikis. They must likewise evade immune cell killing or co-opt immune cells to assist them in completing subsequent steps of the metastatic cascade. And finally, they must resist hydrostatic sheer (i.e., turbulence within vessels).The relative susceptibility to insults can vary widely and may contribute to metastatic inefficiency in a stochastic way. Nonetheless, a successful metastatic cell must have evaded whatever insults to its survival were mounted, whether by chance or by design.

Modulation of the secondary site: Metastatic cells manipulate the tissues that they colonize. For example, in bone, breast and prostate cancers disrupt the balance of osteoclasts and osteoblasts to cause bone resorption or deposition, respectively. Even at a rudimentary level, tumor cells reorganize the extracellular matrix to create a more hospitable environment. Often disruption of homeostasis is the underlying cause of symptoms rather than simply tumor size.

Proliferation at the secondary site: Ultimately, the mere ability to disseminate and arrest at a secondary site is not adequate to constitute a metastasis. Vagrant cells must colonize the secondary site. This notion is key to a definition of metastasis and will be elaborated further below.

These hallmarks would seem to incorporate the criteria necessary to define a metastasis. But, a definition serves an important function - to discriminate two similar, but distinct, things. Importantly, all of these properties must co-exist within a single cell since metastases arise predominantly from single cells (27,28). So, do these hallmarks of metastatic cells discriminate metastatic cells from non-metastatic cells? Surprisingly not.

The ability to invade is not unique to cancer cells. Leukocytes and neurons invade tissues as part of inflammation and normal development, respectively. Similarly, leukocytes and stem cells exhibit intermittent adhesion as part of their normal function. And while moving around the body, they are certainly resistant to anoikis. Each of the cells listed above exerts influence upon the secondary site. During inflammation, for example, leukocytes and fibroblasts degrade and reconstitute extracellular matrix. Proliferation of cells at two different locations would seemingly distinguish metastatic cells from normal counterparts; however, macrophages and stem cells (e.g., angioblasts) can proliferate at secondary sites. Even the latter can persistently proliferate at secondary sites. Together, the distinctions between metastatic tumor cells and normal cells are difficult to identify. Making matters even more challenging, metastatic cells use (essentially) the same mechanisms for accomplishing each of the steps as do their normal counterparts.

Unlike stem cells which can enter a secondary site, proliferate and differentiate, metastatic cells do not differentiate fully at a secondary site. To the best of my knowledge, there is no evidence that stem cells retain pluripotency upon colonization of secondary sites. Hence, another hallmark of metastatic cells is their ability to persistently proliferate without fully differentiating.

Yet, even cells capable of persistently growing at the primary tumor site or in distant tissues are not able to do so indiscriminately. Metastases do not occur randomly (29,30). Depending upon tumor origin, the pattern of colonization varies, dependent upon anatomic distribution patterns, tissue susceptibility to invasion and tissue hospitality. Disseminated tumor cells must

make two critical decisions: (1) whether to begin growing; and (2) whether to continue dividing. The decisions will be based upon a cell's ability to express the full complement of genes required to do so, receptor profile along with corresponding intracellular signaling pathways, level of differentiation at the time of arrival, and the microenvironmental milieu.

As the steps in the metastatic cascade are described, certain principles emerge.

- Metastasis is distinct from tumorigenicity. This principle is demonstrated most directly and elegantly by data demonstrating that certain molecules, known as metastasis suppressors, block metastasis without blocking tumorigenicity (31-33).
- Metastatic potential of individual cells varies between people, by tumor type and within a given tumor (i.e., heterogeneity).
- Metastases arise via multiple routes - hematogenous, lymphatic, across epithelial or serosal cavities.
- The majority of secondary tumors are clonal in origin (34,35). As they proliferate, heterogeneity can (and often does) redevelop (36). Among the earliest characteristics of transformed cells are genetic and phenotypic instability. Cancer cells are more prone to mutation and phenotypic drift than their normal counterparts (37,38). Genetic instability, coupled with Darwinian notions of the 'survival of the fittest', results in selection of populations resistant to host growth control, immune selection, and environmental parochialism (36). Neoplasms tend to become more aggressive with time, but the rate of progression is tumor-dependent and not all cells within a tumor are more aggressive. This latter point is extremely important because, within any neoplastic mass, subpopulations can be isolated with variable behaviors. So, not all cells within 'metastatic tumors' capable of metastasizing (39-43). Even cells isolated from large metastases show heterogeneity in their abilities to metastasize when evaluated experimentally (44), raising questions about whether there is a transiently metastatic compartment (i.e., cells temporarily acquire metastatic capacity (36,45)). Recently, the notion of metastatic cancer stem cells has gained momentum (46). The heterogeneity of metastases with regard to metastatic potential could also be explained by existence of a minority population. While appealing, the actual existence of such cells has still not been definitively demonstrated (47).
- Metastases are non-randomly distributed, invoking tumor-host interactions as key regulators of the process (29,48,49).
- Metastasis is an extraordinarily inefficient process (7).
- Every step in the metastatic cascade is rate-limiting. The inability of a given cell to successfully colonize a secondary site could be due to a deficiency at any step. Examples exist in different tumor models showing failure to metastasize caused by the inability to intravasate (50), survive in the circulation (12,13), survive immune insult (51,52), invade (53), extravasate (54,55) and proliferate at the secondary site (18,56,57). While tumor cells must complete every step of the metastatic cascade, intrinsic deficiencies may be complemented by accompanying cells, whether tumor or host.

Are disseminated emboli metastases?

Any definition of metastasis is probably most controversial when considering the latter two steps - extravasation and proliferation. Extravasation is not required prior to proliferation but proliferating tumor cells eventually leave from the vascular compartment analogous to an over-filled balloon. The question arises: Do proliferating cells within a vessel constitute a metastasis?

According to Leighton (58), the issue is clouded because presence of emboli (i.e., intravascular clusters of tumor cells) is common. He further wrote that,

> "Every pathologist has seen arterioles that are partially or completely occluded with mixtures of tumor cells and fibrin-like material. ... Even so, there may be no evidence of tumor infiltration or extension into the interstitial tissue or alveolar spaces. The importance of this type of finding is to demonstrate that the attachment of a tumor embolus to the wall of a small vessel does not allow us to predict with certainty that it will form a metastasis, a new tumor nodule in the tissue."

Elsewhere in the same volume, Leighton wrote that the notion that intravascular emboli in the lungs as non-metastases had been resolved in the early 20th century. Yet, the issue still persists in current literature. It might be possible to discriminate non-dividing intravascular tumor cells from proliferative ones, but it would require additional stains and assays (e.g., Ki67 immunohistochemistry). In deference to Leighton's assertion, the definition of metastasis will require that at least some of the cells extravasate.

The issue of proliferation at a secondary site is most sticky. In efforts to identify the step(s) of the metastatic cascade inhibited by metastasis suppressors, we and others found (39). cells re-expressing metastasis suppressors were capable of completing every step of the metastatic cascade, except proliferation at the secondary site (33). I will illustrate using an example from my laboratory (18). Briefly, we GFP-tagged metastasis competent C8161 human melanoma cells along with their metastasis suppressed neo6/C8161 counterparts. Following intravenous injection into immunocompromised mice, both cells distributed equally well throughout the body. The number of microscopic foci in various organs diminished equally well over 7 days. However, only the metastatic C8161 cells divided in the lungs and killed the mice within ~4 weeks. In contrast, neo6/C8161 cells were still present in the lungs several months following injection. They were either single cells or clusters of <10 cells. neo6/C8161 cells isolated from the lungs were established in culture. The cells still fluoresced, maintained the resistence markers for the GFP vector and the added chromosome (puromycin and neomycin, respectively), and still retained the entire chromosome (as determined by PCR analysis of markers spanning the entire chromosome). Upon intradermal injection into athymic mice, the cells formed tumors. More importantly, single cells that had disseminated to the lungs could be observed.

Are single disseminated cells metastases?

The answer to this question is not straightforward. If stipulated that millions of cells enter the vasculature and that most do not successfully form macroscopic lesions, then the fate of the non-colonizing cells must be known before a clear definition can be advanced. Since many cells persist at secondary sites without colonizing the tissues, they should hardly qualify as *bona fide* metastases.

The presence of disseminated single cells within distant tissues possibly explains why supposedly "cured" cancer patients recur several years after initial diagnosis and treatment with a "disease-free" interim. Tumor cells had obviously disseminated prior to diagnosis, but remained dormant, or very slowly proliferating, for extended periods. Regardless, the cells that persisted in the distant tissues had ***metastatic potential***—i.e., the capacity to form a macroscopic lesion if circumstances permitted. Apparently, the conditions for division were not suitable during the intervening period.

The presence of single cells (or small finite numbers of cells) with metastatic potential at distant sites has been the subject of considerable interest. TNM and AJCC staging definitions now incorporate aspects of so-called micrometastases (59-61). Based upon their publications, Pantel and colleagues advocate that even single cells are metastases (62). While they provide data

that disseminated single cells in bone marrow or lymph node may be prognostic indicators for some cancers, the mere presence of tumor cells does not constitute a metastasis. Unfortunately, examination for microscopic metastases is not systematic, except in regional lymph nodes and, occasionally, bone marrow. The critical question is whether one can discriminate patients in whom disseminated cells will remain indolent from those who will eventually progress to overt disease. The answer will allow patients who have disseminated cells with little likelihood of developing overt metastases to avoid the ravages of cytotoxic therapy.

How to accurately predict outcome is the most important advance that will impact cancer cure rates. Toward that end, several laboratories are utilizing expression microarrays to identify metastasis 'signatures.' Such studies have the potential to revolutionize the practice of pathology, bringing a molecular answer to the vexing question. Van't Veer and colleagues used a 70 gene set to identify and define a 'poor prognosis' transcriptome in breast cancer (63), which was used to predict survival (64). The ability of microarray data (and proteomic data as well, although not the subject of the papers cited) to stratify 'at risk' patients from those with lesser risk is not questioned. However, the use of microarray data to ascribe function and mechanism is challenged (28). Briefly, microarrays comparing primary tumors and metastases from multiple tumor types have shown remarkably similar patterns (65,66). Unfortunately, the interpretation that metastatic potential is 'hardwired' into tumor cells does not hold when experimental methods are scrutinized (28,31). First, microarrays were done using samples 'contaminated' with stromal cells. Since arrays of normal tissues from mice with susceptibility or resistance to metastasis yield similar metastasis signatures (67,68), then interpretation is not straightforward. Second, samples contained RNA from mixtures of tumor cells. We know that metastases arise from single cells with all requisite expression patterns. Merely finding expression within a large neoplastic mass does not provide any information regarding whether all of the genes are coordinately expressed within a single cell. However, the use of expression arrays as adjuncts to other tools shows great promise for decreasing the subjectivity mentioned above (69).

If one again stipulates that tumor cells disseminate at relatively high frequency when a primary tumor is present and that the proportion of disseminated cells eventually forming macroscopic lesions is a fraction of the cells entering the circulation, it follows that the distinctive trait of metastatic cells is the ability to proliferate at the secondary site. As early as 1919, James Ewing referred to an uncited study by M. Schmidt who said, "From the study of lung in these cases [of cancer], Schmidt concludes that in cancers of the abdominal organs, there is frequent and repeatedly a discharge of cancer cells which lodge in the small arteries of the lungs [but fail to form metastases]."

Yet, presently it is not known whether the persistent disseminated cells are dormant (i.e., non-dividing) or are of limited replication ability (i.e., balanced division and cell loss; rare division). The former would explain resistance to prior therapies that, for the most part, target proliferating cells. The latter would provide a potential mechanism for new mutations which become passed along to progeny (e.g., loss of metastasis suppressor function). Conversion to a fully proliferating cell could also occur in response to changes in the host tissue (e.g., injury, change in hormonal status, aging). The mechanism(s) responsible for acquisition to proliferative potential do not necessarily have to be known in order to define a metastasis.

Nonetheless, Willis wrote that, "only those ***growths*** [emphasis added] which are separate from the primary growth and have arisen from detached transported fragments of it [the primary tumor] are entitled to be called 'metastases (70), ." He then elaborates regarding the discontinuity of metastases from the primary tumor so that his point regarding growth is somewhat obfuscated. Both points are critical.

Growth was not an issue until techniques capable of detecting single cells or emboli were developed. Indeed, happenstance sections in tissues often detect disseminated neoplastic cells. But their clinical importance was negligible since they were not impacting function. Microscopic lesions seldom alter tissue functions and are seldom lethal themselves. This does not minimize the potential danger caused by disseminated cells since they have accomplished the majority of antecedent steps in metastasis. In a sense, they are analogous to terrorist cells waiting to be activated. The potential for destruction is present. But when they blend innocuously, they are difficult to detect and to neutralize. The challenge is to discriminate between those cells merely leaning toward destructive behavior from those truly committed to killing the host.

Failure to distinguish *bona fide* metastases from disseminated cells has some important implications. Current medical practice is to eliminate all risk. Therefore, if cells have already spread, then aggressive treatments are advocated. Additionally, the extent of spread and the location of spread (i.e., local *vs.* regional *vs*. distant) determine the treatment plan. This is perfectly logical. The problem is that patients may be subjected to more cytotoxins than necessary since the majority of disseminated cells fail to colonize secondary sites. It is colonization that directly puts the patient at risk, not merely the potential to do so.

At an experimental level, the implications are no less important. There are literally hundreds of manuscripts published yearly where tumor cells are introduced into the vasculature and in which single cells are detected in the lungs (or other organs) and called metastases. If, however, the animals were not euthanized prematurely and were maintained for long periods, they never developed overt metastases. Can one then call the tumor cells metastatic? Absolutely not!

A definition of metastasis

Taking the above considerations into account, I would like to propose the following definition of metastasis:

> The dissemination of neoplastic cells to discontiguous nearby or distant secondary (or higher order) sites where they proliferate to form an extravascular mass of incompletely differentiated cells.

This definition includes an implicit requirement for a primary tumor, even if one cannot be detected or located. Metastases, following upon Willis' criterion, are not direct extensions of the primary tumor, even if extended over long distances. The route of dissemination does not determine whether a secondary lesion is a metastasis, but at least some of the cells must no longer remain within a vascular compartment. There is question about the necessity for tumor cell extravasation prior to proliferation. The point is moot. Proliferation can begin within a vessel and continue, or division can be initiated after cells have extravasated. In either case, Leighton's cautionary tone regarding intravascular cells warranted inclusion of a criterion for whole or partial extravasation.

There can also be metastases from metastases. Owing largely to the inability to detect asymptomatic microscopic lesions, the natural progression of tumors in humans is extrapolated rather than directly measured. Based upon incidence data, some would argue that most epithelial tumors spread first to regional lymph nodes. Such staged tumor cells can then migrate to the blood stream for more widespread dissemination. Interestingly, however, breast and prostate cancers metastasize more frequently to bone than to lymph nodes. Still, the concept that tumor cells can emigrate more than once is not questioned.

The most difficult parameter to define is *macroscopic*. With continued improvements in technology, the ability to detect smaller foci will continue to increase. As little as five years ago, bone metastases were defined by X-rays showing osteolysis or osteopetrosis. Within the

last year, we have been able to identify, in a laboratory setting, single cells inside bone using GFP-tagged cells (71,72). Equally astonishing improvements have been made with microcomputerized tomography in both the clinical and laboratory realms. Therefore, resolution may one day be at the level of a single cell and clinical decisions will need to be made on that basis. However, the distinction between single disseminated cells and overt metastases should remain.

To partially resolve the complexity regarding what constitutes a macroscopic lesion, I will use criteria used by radiation biologists faced with a similarly vexing issue when studying cellular response to radiation. Briefly, many cells have limited proliferative potential following radiation treatment. In other words, they can divide a couple times before dying or becoming quiescent. Early radiobiologists questioned the threshold of colony size necessary to consider survival (73,74). Similar issues were raised during early studies of clonogenicity in soft agar (75). Although the exact numbers vary slightly, five to six cell divisions are typically required to qualify as a colony ($2^6 = 64$ cells). Therefore, I propose that a metastasis must constitute at least 50 cells.

Implications

Ultimately, all cancer treatment besides resection of the primary tumor is targeted to the prevention of metastasis or the elimination of already-established metastases. Current staging systems focus on primary tumor size and local invasiveness in addition to lymph node status and the presence/absence of distant metastases. Thus, the TNM and AJCC staging systems already have two parameters directly related to metastases. The reason, ostensibly, is that lymph node metastases provide different clinical information from visceral metastases. If one extrapolates these assumptions to a logical end, the TNM system (and variations) may need to be refined in order to take into account not only the presence/absence of disseminated cells (i.e., metastatic potential), but also location and size of metastases.

Segregation of lymph nodal involvement should be extended to define the location of metastases throughout the body. A system which merely has binary input for metastasis is inadequate. Location is important because in patients with melanoma, for example, metastases to the subcutis are better off than patients with metastases in the brain (76,77). The TNM and AJCC staging systems have already begun to evolve in this direction (59,60). M1(Hep), M1 (Pul) and M1(Oss) have been incorporated to describe hepatic, pulmonary and osseous metastases (76,78). Moreover, non-traditional staging and clinical group issues have been incorporated as well and show promise.

As sentinel lymph node studies are already coming to grips with the issue (79), the size of disseminated cell clusters has become a critical parameter. Similarly, patients with smaller metastases are typically in lesser immediate danger than those with larger metastases. Current recommendations define microscopic metastases as clusters of cells <0.2 mm in size (indicated by N0(i) to indicated isolated tumor cells) (59,60). This size partially addresses a minimal mass issue outlined above since this would represent approximately 8-10 cell diameters in two dimensions.

The TNM and AJCC staging systems are already evolving to incorporate elements of location and size. But even more attention is needed. Therefore, I would like to propose a TLS system (**T**umor, **L**ocation of metastases, **S**ize of metastases). Tumor stratifications would parallel those of the current TNM/AJCC systems. Location would be more extensive than the node/distant metastasis mechanisms currently utilized. And size would come into play for each metastasis at each location. I suspect that the size category may be stratified by methodology as well as physical measurement. For obvious reasons, changeover would be difficult and should not be undertaken capriciously. However, the benefits gained by accuracy and presumed decision-

making ability would be worth the effort. Results from the laboratory and clinic would benefit by a single, clear definition of metastasis that transcends models. The first step toward this end is consensus regarding what constitutes a metastasis.

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