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DIAGNOSTIC MORPHOMETRY: AIMS, TYPES, CONDITIONS AND APPLICATIONS

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ABSTRACT

Two general types of quantitative pathology applications are available for routine use: image processing and flow-cytometry. This paper deals with a subset of image processing in which most of the diagnostic applications available today are found: diagnostic morphometry.

The aims of quantitative pathology are to eliminate the subjective nature of the usual cytological and histological evaluations, which imply a considerable degree of disagreement between pathologists. Secondly, the quantitative and continuous nature of morphometrical features allows the expression of the numerical classification probability. Automatic classifiers with digital image processing computers at the moment are not (yet) available and therefore, the pathologist has to select the diagnostically interesting cells and areas. Thus, it is a SELECTIVE approach, which together with multivariate analysis is an essential condition in the useful application of diagnostic morphometry today.

The applications so far available can be subdivided in three types: for quality control, to support or to decide for a certain diagnosis or grade, or to decide for a certain treatment. Obviously an increasing degree of reliability is required from the first to the third type. In addition, five levels of quality can be discerned in quantitative pathology studies. First level studies describe discriminating features (to distinguish two or more groups). A multivariate classification rule with numerical classification probabilities are the highest quality within this level. Level II studies confirm the results of level I studies in a test set, in the same laboratory. Level III studies confirm the results of level I studies in a test set, but now in another laboratory, on different material by independent investigators. Alternatively the same results may be found independently in several laboratories in independent studies. Level IV studies confirm level I findings, when routinely used in the same laboratory. Level V studies are similar to level IV studies, but now in different laboratories.

Most of the applications so far available are type 1 or 2, level I-III. A few applications are level IV and the number of level V applications is re-

stricted. In contrast, level I-III studies are rapidly increasing.

Although further multicenter studies are essential, diagnostic morphometry applications so far available confirm the original expectations that this method is useful in diagnostic pathology. Several of the original aims already have been reached. Further development, especially in the area of digital image processing is a promising future possibility.

INTRODUCTION

Routine assessments of histological and cytological diagnoses contain a considerable degree of subjectivity. In a research setting, quantitative microscopical techniques have been used for more than 50 years, but diagnostic quantitative microscopical applications started to enter pathology laboratories approximately 1 decade ago. At the beginning of the 80's, the number of applications sharply increases as is also clear from the establishment of two journals devoted to quantitative microscopy in pathology (Analytical and Quantitative Cytology; Cytometry). No doubt, the outstanding pioneer work by Caspersson in the 30's and the basic work in quantitative cytology by Wied, Bahr, Bartels, Koss, Young and others in the United States in the 60's and 70's have exerted a considerable influence in this direction. Interactive applications, using stereology so far mainly are developed and used in Europe,most likely under the influence of the Swiss anatomist Weibel.

Quantitative pathology today includes two general types: image processing and flow-cytometry. For an overview of the latter, reference is made to Baak and Oort (1983) and Herman et al (1984). Image processing applications of quantitative pathology can be subdivided in interactive and automatic methods. Interactive methods include diagnostic morphometry and DNA-ploidy measurements by slide scanning. This paper is restricted to diagnostic morphometry and the following points will be discussed: the aims, types, conditions and levels of applications. Finally some new applications will briefly be summarized.

AIMS

A variety of arguments applicable to quantitative techniques are connected with objectivity (Editorial, 1976) and reproducibility. In medical disciplines other than pathology, quantitative assessments have already been in use for a long time (for example: haematology and clinical chemistry). Apart from efficiency reasons lack of consistency with subjective methods was an important reason for this.

Pathologists usually agree very well, both with themselves and with each other. In several diagnostic areas there is some or considerable disagreement, especially in the so-called continuous type of pathological lesions (see Langley et al, 1983). Most of these lesions are seen in tumour pathology and differences in for example tumour grade can result in differences in therapy. This explains the need for more objective and more consistent methods. Due to its quantitative nature, morphometry fulfills these demands. In addition, the continuous nature of the quantitative features makes them especially accessible to assess a certain diagnosis or grade in a quantitative way. In his outstanding study, Toogood (1981) shows that the same words

used by different pathologists contain a considerable difference in meaning, and this confirmed earlier findings by Bryant and Norman (1980). For example, the term "probably malignant" for one pathologist means "in my opinion there is invasive growth or a metastasis somewhere, although I do not see these features in the present section". For another pathologist these words mean "it is an atypical lesion and it has some, but not all the morphological features of malignancy". The clinician interpretes these formulations in his own way, and a variety of therapeutic measurements may result (Toogood, 1981). Numerical classification probabilities, calculated for a combination of two, three or more features overcome this problem (Baak et al, 1983).Thus, the agreement between different pathologists can be improved in this way.

Another advantage is, that elements, compartments and other cytological or histological features have to be defined much more strictly, in order to measure them accurately. This in itself results in an improvement of the quality.

TYPES

Three types of diagnostic morphometry applications can be discerned:

1. Quality control applications;
2. To support or to decide for a certain diagnosis or grade;
3. To decide for a certain therapy.

Apparently, a higher degree of quality and reliability of the methods is required going from the first to the third type of applications. Type 3 applications require very high quality and consistency; a low coefficient of variation of the methods and assessments are a prerequisite for this.

CONDITIONS

The complexity of cell and tissue images used has sofar prevented to perform the quantitative analysis fully automatically. Quantitation still is possible as an interactive semi-automatic method, in which the pathologist selects objects to be measured. This approach is called "interactive diagnostic morphometry". That such an approach is much more reproducible and consistent than subjective evaluation is caused by the phenomenon of "simultaneous contrast" (Cornsweet, 1970) - i.e. different areas within the same tissue section are simultaneously seen and compared with each other and selected for measurement.

This in fact is the same as in usual subjective evaluation. However, the features then are quantitatively described instead of subjectively graded. Here the difference begins. Subsequently, comparison with the typical images in the computer memory, instead of the human mind and classification follows.

Again, these two latter steps are completely stable with morphometry, whereas with subjective evaluation they are subject to many distracting psychophysiological factors. However, the first basic step (selection of the suitable areas, cells) should be done properly. Thus, outstanding skill of the pathologist is an essential condition for useful interactive diagnostic morphometry application.

A second condition, which is perhaps equally important, is that multivariate analysis should be applied. Single quantitative variables usual-

ly show a considerable overlap, which in the past has disappointed many pathologists. However, the combination of, each for itself, overlapping features may be completely discriminating, as is clear from many studies. For a more detailed description of this reference is made to Baak et al, 1983.

A third condition is that only features are used which are not, or hardly, influenced by possible variations in tissue or cytoprocessing techniques. Interestingly enough, the majority of the diagnostically relevant features fulfill this demand whereas more complicated quantitative microscopical characteristics (such as chromatin pattern descriptors) are much more sensitive to slight variations (see Velthuis et al, 1978; Thunnissen et al, 1981).

APPLICATIONS

Although the increasing interest and number of articles describing diagnostic morphometry applications is encouraging, there is a certain risk that the quality of the different studies differs. For actual application of morphometry in individual patient care several precautions have to be taken, and different levels of quality have to be discerned within the studies.

Five different levels can be discerned:

Level I. Studies which describe quantitative features,either single or in combination, to distinguish two or more diagnostically or prognostically different groups.The decision threshold(s) should be adequately described, together with sensitivity, specificity and efficiency.

Level II. Confirmation of level I findings in an independent testset of patients or cases, obtained from the same laboratory. This minimizes the risk of classification shifts due to variations in tissue processing and measurements (see section: Conditions).

Level III. Confirmation of level I findings in a testset in an independent laboratory. This makes the findings much more impressive, as the discriminating features apparently are independent of interlaboratory variations and interobserver measurement differences. As an alternative, different independent level I or II studies in several laboratories obtaining the same results and conclusions, belong to this level.

Level IV. Confirmation of level I-III findings when routinely used in the same laboratory. Routine application of a technique usually results in a somewhat lower quality. If the same results still are obtained, it again emphasizes the "strength" of the diagnostic morphometrical classifier.

Level V. Confirmation of level I-IV findings when routinely used in other independent laboratories.

Very few of the applications so far described have reached level V. Some are level IV studies, but many investigations still are restricted to level I studies. Especially if such articles only describe the means of two groups, and the probability of no difference ("significance"), although formally level I, they are nearly useless for diagnostic applications. Editors of journals should require the authors of such articles to describe their results properly, indicating the decision threshold for a feature (or features when a combination is used), and to express their findings in terms of sensitivity, specificity and efficiency.

For an overview of applications in tumour pathology and non-tumour pathology, reference is made to chapter 6 and 7 of the Manual of Morphometry in Diagnostic Pathology (Baak and Oort, 1983). Several new classifiers have been developed and described since the publication of this book and many more can be expected in the near future. Other, somewhat older applications have considerably been improved, and these publications now also are in preparation. The prediction of the prognosis of malignant melanoma, positivity or negativity of OR in breast cancer, prediction of the prognosis of ovarian tumours and distinction of undifferentiated adenocarcinoma and granulosa cell tumours of the ovary are a few examples. Prostate morphometry is well in progress, kidney morphometry has been expanded in the past two years also. the usefulness of morphometry of malignant lymphomas, especially mycosis fungoides and Sezary syndrome, has been confirmed in several independent studies, which makes this to a level III application. Several independent studies have confirmed the usefulness of breast cancer morphometry for the prediction of the prognosis and this method has been used routinely for four years in our laboratory, as well as endometrial and ovarian tumour grading with morphometry. The results of this partly have been published. They can be regarded as level IV applications.

Further introduction of digital image processing computers and DNA-ploidy measurements are important and promising future possibilities.

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