Transcontinental communication and quantitative digital histopathology via the Internet; with special reference to prostate neoplasia

R Montironi, D Thompson, M Scarpelli, H G Bartels, P W Hamilton, V D da Silva, W A Sakr, B Weyn, A Van Daele, P H Bartels

Objective: To describe practical experiences in the sharing of very large digital data bases of histopathological imagery via the Internet, by investigators working in Europe, North America, and South America.

Materials: Experiences derived from medium power (sampling density 2.4 pixels/µm) and high power (6 pixels/µm) imagery of prostatic tissues, skin shave biopsies, breast lesions, endometrial sections, and colonic lesions. Most of the data included in this paper were from prostate. In particular, 1168 histological images of normal prostate, high grade prostatic intraepithelial neoplasia (PIN), and prostate cancer (PCA) were recorded, archived in an image format developed at the Optical Sciences Center (OSC), University of Arizona, and transmitted to Ancona, Italy, as JPEG (joint photographic experts group) files. Images were downloaded for review using the Internet application FTP (file transfer protocol). The images were then sent from Ancona to other laboratories for additional histopathological review and quantitative analyses. They were viewed using Adobe Photoshop, Paint Shop Pro, and Imaging for Windows. For karyometric analysis full resolution imagery was used, whereas histometric analyses were carried out on JPEG imagery also.

Results: The three applications of the telecommunication system were remote histopathological assessment, remote data acquisition, and selection of material. Typical data volumes for each project ranged from 120 megabytes to one gigabyte, and transmission times were usually less than one hour. There were only negligible transmission errors, and no problem in efficient communication, although real time communication was an exception, because of the time zone differences. As far as the remote histopathological assessment of the prostate was concerned, agreement between the pathologist’s electronic diagnosis and the diagnostic label applied to the images by the recording scientist was present in 96.8% of instances. When these images were forwarded to two pathologists, the level of concordance with the reviewing pathologist who originally downloaded the files from Tucson was as high as 97.2% and 98.0%. Initial results of studies made by researchers belonging to our group but located in others laboratories showed the feasibility of making quantitative analysis on the same images.

Conclusions: These experiences show that diagnostic teleconsultation and quantitative image analyses via the Internet are not only feasible, but practical, and allow a close collaboration between researchers widely separated by geographical distance and analytical resources.

Digital image technology (DIT) has several applications in pathology, including telepathology, teaching, and quantitative digital histopathology. Research in quantitative histopathology tends to be directed at problems where visual diagnostic assessment has noticeable interobserver variability—for example, the evaluation of preinvasive epithelial neoplasms of the breast, endometrial lesions, and prostatic intraepithelial neoplasia (PIN) lesions. Further applications are in areas where very early changes can be detected, such as changes that cannot be detected unequivocally by visual examination alone. In addition, there are applications where small changes—in lesion progression or regression as a result of chemopreventive intervention—need to be documented quantitatively. Diagnostic expertise for these difficult problems in histopathology tends not to be generally accessible.

Quantitative histopathology demands substantial logistic support, in microphotometric equipment, computer resources, software, and human diagnostic and analytical expertise. In a histometry assessment, a representative region of a histopathological section may require the recording, processing, and analysis of hundreds of video frames. Interactive correction of segmentation is no longer practical. Knowledge guided segmentation software for an automated and correct segmentation of the complex imagery offered by histopathological sections represents a major software development.

Abbreviations: DIT, digital image technology; FTP, file transfer protocol; JPEG, joint photographic experts group; LAN, local area network; NP, normal appearing prostate; NPadj, normal appearing adjacent to prostate cancer; Npdist, normal appearing distant from cancer; OSC, Optical Sciences Center, Arizona, USA; PCA, prostate cancer; PIN, prostatic intraepithelial neoplasia; POTS, plain old telephone service; ISDN, integrated services digital network; TIFF, tagged image file format.

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Table 1 Institutions and persons involved in projects related to prostate neoplasia, in addition to the tasks more specifically carried out in the different laboratories

<table>
<thead>
<tr>
<th>Laboratory</th>
<th>Institution/Country</th>
<th>Contact person(s)</th>
<th>Main task(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>(1) Image Analysis Laboratory</td>
<td>University of Arizona, Tucson, Arizona, USA</td>
<td>Peter H Bartels, Deborah Thompson, Hubert Bartels</td>
<td>Software/hardware development, image analysis, statistics</td>
</tr>
<tr>
<td>(2) Institute of Pathological Anatomy</td>
<td>University of Ancona, Ancona, Italy</td>
<td>Rodolfo Montironi, Marina Scarpelli, Peter W Hamilton</td>
<td>Pathology consultation, slide provider, immunohistochemistry</td>
</tr>
<tr>
<td>(3) Laboratory of Image Analysis</td>
<td>The Queen's University, Belfast, Northern Ireland, UK</td>
<td>Vinicius Duval da Silva</td>
<td>Diagnostic decision support systems</td>
</tr>
<tr>
<td>(4) Department of Pathology</td>
<td>Federal University of Rio Grande do Sul, Porto Alegre, Brazil</td>
<td>Willem Jacob, Barbara Weyn, André Van Doele</td>
<td>Image analysis</td>
</tr>
<tr>
<td>(5) Center of Electron Microscopy</td>
<td>University of Antwerp, Antwerp, Belgium</td>
<td>David S Alberts, James R Marshall</td>
<td>Consultation, image recording</td>
</tr>
<tr>
<td>(6) Laboratory of Image Analysis</td>
<td>Arizona Cancer Center, University of Arizona, Tucson, Arizona, USA</td>
<td>Waal A Sakr</td>
<td>Consultation, quality control</td>
</tr>
<tr>
<td>(7) Department of Pathology</td>
<td>Wayne State University, Detroit, Michigan, USA</td>
<td>James R Marshall, David S Alberts, André Van Doele</td>
<td>Consultation, image recording</td>
</tr>
</tbody>
</table>

This project gave rise to a practical problem.

The technical personnel—who had no formal training in histopathology—recording the imagery needed guidance in the selection of areas to be scanned and in the identification of areas of normal prostate, high grade PIN, and PCa. All recorded fields had to be verified and accepted by the remote diagnostician (remote histopathological assessment).

The personnel recording the images had certain rules to follow, namely:

1. In each slide, when possible, normal looking ducts/acini adjacent to and distant from cancer as well as high grade PIN and PCa have to be imaged.
2. Complete ducts/acini featuring normal looking tissue and high grade PIN have to be present in each image. For PCa, the image has to be large enough to allow determination of the primary Gleason grade.
3. In each case, several ducts and acini of normal prostate and high grade PIN and several areas of PCa have to be recorded.

The clinical material was provided by the department of pathology, Harper Hospital, Wayne State University, Detroit, Michigan, USA (Dr W Sakr). It consisted of 75 radical prostatectomy specimens with prostate cancer of the peripheral zone. None of the patients had received chemotherapy, hormone treatment, or radiotherapy before surgery. In each case, the slide with the largest focus of PCa, also containing high grade PIN and normal prostate tissue, was selected for the investigation.

Immunohistochemistry was performed at the Institute of Pathological Anatomy, University of Ancona School of Medicine, Ancona, Italy, on 5 µm thick paraffin wax embedded tissue sections. The details of the technique have been reported in a previous publication. Briefly, the basal cell layer was immunostained with a monoclonal antibody directed against high molecular weight keratin (34BE12 antikeratin monoclonal antibody provided by Dako Spa, Milan, Italy). The sections were counterstained with a light haematoxylin and eosin stain. The basal cell layer appeared dark brown because of the immunohistochemical detection method, whereas the secretory cells showed a slightly eosinophilic cytoplasm and a basophilic nucleus. This immunohistochemical approach was adopted to facilitate the scientist doing image recording and analysis in the identification of the secretory cell type in normal looking epithelium and PIN and in the identification of the cancer cells.

Histological images were recorded at the image analysis laboratory of the OSC. The imagery was transmitted via the Internet from Tucson to Ancona, Italy. For each image the accuracy of the diagnostic labelling of the lesion or tissue was ascertained before any quantitative image analysis was begun. This type of teleconsultation is called static telepathology.
Hardware and software components used in the remote histopathological assessment

Image acquisition and compression

Image acquisition was carried out at the Optical Sciences Center, University of Arizona, Tucson, Arizona, USA. A Sun workstation was used. The images were recorded on a Leitz videophotometer (Wetzlar, Germany) equipped with computer controlled coordinate stage with a 0.1 µm accuracy. A 20 : 1 Nikon plan apochromatic objective of 0.75 numerical aperture (Melville, New York, USA) was used. A Sony DXC-760 MD three chip CCD colour camera (Melville) was used to record and digitise images. The spectral bands for the red, green, and blue channels were centred at 600 nm, 540 nm, and 460 nm, respectively. The resulting sampling was of 2.4 pixels/µm. Each 24 bit colour digitised frame was 512 by 470 pixels in size requiring 721,920 kilobytes (kB) of disk storage (1 byte = 8 bits).

Frequently, individual digitised frames obtained with the videophotometer with a 20 : 1 objective did not include the full extent of the histological structures. For instance, a duct with high grade PIN features can extend over multiple camera fields. Therefore, it was necessary to record adjacent frames (fig 1A,B) and to merge them in software (fig 1C). This was done by using best fit correlation techniques to align the adjacent frames and optical density ramp functions to blend the strip of overlap between them. For some lesions, the merging process could be extended over a large area, forming multi-megapixel arrays.

The images were then stored in a format developed at the OSC (“ima”). The same images were also converted to TIFF (tagged image file format), which is a format that does not compromise image quality and is suitable for image analysis on PC and Mac computers. There are no differences in detail or quality between these two file formats. For instance, the image shown in fig 1C has a dimension of 976 by 461 pixels (“ima” native format or TIFF format; 24 bit colour depth; size, 1.35 megabytes (MB; 1 MB is equal to 1024 kB)). However, such image sizes are impractical for Internet transmission. Therefore the images were converted to JPEG (joint photographic experts group) using a set compression ratio. JPEG is considered the standard for storing high resolution photographic images, has universal multiplatform support on web browsers.

<table>
<thead>
<tr>
<th>Diagnostic category</th>
<th>No. of images</th>
<th>Total size (MB)</th>
</tr>
</thead>
<tbody>
<tr>
<td>(1) Normal prostate adjacent to PCa</td>
<td>220</td>
<td>47</td>
</tr>
<tr>
<td>(2) Normal prostate distant from PCa</td>
<td>367</td>
<td>66</td>
</tr>
<tr>
<td>(3) High grade prostatic intraepithelial neoplasia</td>
<td>397</td>
<td>78</td>
</tr>
<tr>
<td>(4) Prostate cancer (PCa)</td>
<td>183</td>
<td>27</td>
</tr>
<tr>
<td>All groups</td>
<td>1167</td>
<td>215</td>
</tr>
</tbody>
</table>
and graphics editing software, and is the dominant format for still image compression. JPEG offers a host of configurable compression ratios. When an image is converted from TIFF to JPEG, the final file size depends on the degree of compression. The quality of the converted image is inversely related to the compression ratio. When the highest quality (100) was chosen, the size of the image shown in fig 1C was 476 kB. When lower quality levels (75, 50, and 25) were selected, then the sizes decreased (97 kB, 67 kB, and 44 kB, respectively). The default ratio of the image conversion software at the OSC is set at 75. This gave us the largest trade off between size versus quality. JPEG images are used solely for observation purposes or for image scene analysis that has low image quality criteria.

Table 3  Comparison between the diagnostic label and the electronic diagnosis (this comparison does not include the nine cases where the image showed artifacts)

<table>
<thead>
<tr>
<th>Diagnostic label</th>
<th>NP</th>
<th>High grade PIN</th>
<th>PCa</th>
<th>Others</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>NP</td>
<td>568</td>
<td>0</td>
<td>0</td>
<td>13</td>
<td>581</td>
</tr>
<tr>
<td>High grade PIN</td>
<td>0</td>
<td>375</td>
<td>1</td>
<td>19</td>
<td>395</td>
</tr>
<tr>
<td>PCa</td>
<td>0</td>
<td>1</td>
<td>175</td>
<td>6</td>
<td>182</td>
</tr>
<tr>
<td>Total</td>
<td>568</td>
<td>376</td>
<td>176</td>
<td>38</td>
<td>1,158</td>
</tr>
</tbody>
</table>

Agreement: (568 + 376 + 176)/1158 × 100 = 96.6%. “Others” refers to diagnostic entities other than those proposed by the recording scientist. NP, normal appearing prostate; PCa, prostate cancer; PIN, prostatic intraepithelial neoplasia.

Table 4  Reasons for discordance between the diagnostic label and the electronic diagnosis

<table>
<thead>
<tr>
<th>Discordance</th>
<th>No. of cases (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Class I (field selection)</td>
<td>5 out of 1,158 (0.4%)</td>
</tr>
<tr>
<td>Class II (morphological interpretation)</td>
<td>35 out of 1,158 (3.0%)</td>
</tr>
<tr>
<td>Class III (video image quality)</td>
<td>9 out of 1,167 (0.8%)</td>
</tr>
</tbody>
</table>

Image viewing and transfer
The images referred to in our paper were viewed by three different pathologists, each using a different computer. Each computer, although not state of the art today, is adequately equipped to provide good spatial resolution on the monitors (1024 × 768, 1152 × 870, and 1280 × 960). Two monitors were 20 inches in size and one was 17 inches. The video cards for each computer are capable of displaying 24 bit colour. See appendix A for complete details.

Figure 2  [A–E] The successive steps in the computer assisted identification of the spatial positioning of the cells by syntactic structure analysis in a prostate duct/acinus.
The images were evaluated at the Institute of Pathological Anatomy of the University of Ancona by two of the co-authors (RM and MS) and at the image analysis laboratory of a third co-author (VDdS). The Institute of Pathological Anatomy uses a local area network (LAN) to link its computers and data processing devices. The LAN is a high speed communications system with two different types of connection to the Ancona University Internet Service Provider (unian.it): plain old telephone service (POTS) and integrated services digital network (ISDN). To transfer the images, the reviewing pathologists used the Internet application file transfer protocol (FTP) (software: Fetch 3.0.3; Dartmouth Software Development, Hanover, New Hampshire, USA). Image files could be viewed using Adobe Photoshop 5.0.2 (Adobe Systems, San Jose, California, USA), and Paint Shop Pro (Jask Software, Eden Prairie, Minnesota, USA).

Results

The three applications of the telecommunication system investigated were remote histopathological assessment, remote data acquisition, and selection of material.

Remote histopathological assessment

In connection with the quantitative assessment of high grade PIN lesions, a total of 1167 images were downloaded during 10 different online sessions. From the evaluation point of view, each image was considered as a single case. The recording scientist sent 220 images labelled as normal appearing adjacent to prostate cancer (NPadj) and 367 images labelled as normal appearing distant from cancer (NPdist). There were 397 and 183 images sent for high grade PIN and PCa, respectively (table 2).

A minimal number of transmission errors occurred, but a second attempt on those images was successful. Nine images were out of focus so as to preclude their evaluation. They were excluded from the following discussion, leaving 217 images of NPadj, 364 images of NPdist, 395 images of high grade PIN, and 182 images of PCa, making a total of 1158 images.

Table 3 shows the comparison between the pathologist’s electronic diagnosis and the diagnostic label applied to the images by the recording scientist. There was agreement in 96.6% of instances. Table 4 reports the reasons for discordance between the diagnostic label and the electronic diagnosis.

Intraobserver reproducibility

The reviewing pathologist assessed his intraobserver reproducibility in 200 consecutively transmitted images of normal prostate (60 cases), high grade PIN (85 cases), and PCa (55 cases). These cases were re-evaluated three months after they had been received and compared with the diagnostic label. An overall concordance of 98.4% was seen. In particular, a 100% concordance was obtained for normal prostate and PCa, whereas that related to PIN was 95.3%. The discordance in this last group was the result of four cases that originally were considered high grade PIN and later as low grade PIN.

Interobserver reproducibility

One hundred consecutively downloaded images were sent to one of the co-authors (MS) with the LAN (appendix A). Agreement with the electronic diagnosis made by RM was seen in 98.0% of images.

One hundred and five images were sent to the image analysis laboratory of one of the co-authors (VDdS) (appendix A). The material was sent as a file attachment by email (provider’s name: doctormail.com). Email is generally not satisfactory for sending large amounts of data because of the limitations placed on the system in terms of the maximum sizes of incoming mail messages. VDdS considered the material of sufficient quality to allow histological identification of the contents. The agreement between the electronic diagnosis made in Ancona and that rendered in the Brazilian centre was as high as 97.2%.

Remote analytical software

To take advantage of the availability of software providing graph based scene decomposition at the University of Antwerp, Belgium, 100 images were sent to the Centre of Electron Microscopy, University of Antwerp (BW). Images
were sent in groups of 20 as a file attachment by email. The purpose of this study was to apply syntactic structure analysis to quantify the spatial positioning of the cells near the basal layer. This was done by connecting the cells with three different syntactic structure graphs (the Voronoi diagram, Gabriel’s graph, and the minimum spanning tree). These graphs, shown in fig 2A–E, were quantified, resulting in several features that can be put into a classifier to find the most discriminant combination (data not shown).

One hundred images were “ftp-ed” to the image analysis laboratory of the Queen’s University, Belfast, UK. The purpose was to see whether any kind of histometric analysis was possible on the material originally received from the OSC. Figure 3A–C shows the digitised image and the derived images in which the secretory and basal cell nuclei, in addition to the basal cell layer, are identified. Histometric features related to the size of the gaps present in the basal cell layer were evaluated (data not shown).

Remote data acquisition
Locally, at the University of Arizona, all digitised imagery recorded at the Arizona Cancer Center is sent routinely via the Internet to the OSC for analysis. Protocols from the past several months list imagery recorded in a micromorphometric assessment of the effects of vitamin A, at different doses. One such study involved 0.89 GB of image data recorded over a period of one month, at 25 hours each week, and transmitted within less than one hour. Another data set for a different dose of vitamin A involved 0.85 GB. A morphometric control data set for skin shave biopsies to be tested with biomarkers involved another 0.36 GB. Data from a study of endometrial lesions resulted in 0.45 GB and a study of premalignant and malignant breast lesions produced 0.45 GB of image data. Each data set was transferred to the OSC via the Internet and was archived and analysed there (data not shown).

Selection of material
The teleconsultation on very large scale digital imagery was also used to select the areas of the tissue sections to be analysed. The selection and the subsequent analyses were done on medium power (sampling density 2.4 pixels/µm) and high power (6 pixels/µm) imagery, respectively. Frequently, this involved delineation of regions of interest in medium power imagery, to be recorded under oil immersion for karyometry. The following...
are some situations in which the advantages of using teleconsultation in the selection of the material were seen:

- Outlining regions of interest—for example, those representing low grade PIN, high grade PIN, or histologically normal appearing prostate glandular tissue, at a predetermined distance from a PIN lesion. Figure 4 shows an example. Measurements of randomly selected nuclei are then taken only in the outlined region.

- Labelling of nuclei to be recorded under oil immersion in instances when random selection is not advised. Figure 5A,B shows this procedure.

- On occasion, guidance from a remote co-investigator is needed in the visual interpretation of imagery. A good example is a study aimed at testing the heterogeneity of secretory cell nuclei in high grade PIN lesions. The question that arose was what constitutes a true “gap” in the basal cell layer. A set of example images was sent to clarify the situation for the image recording personnel. Figure 6 indicates an area with intact basal cells and an area where the basal cells are not present.

- In studies on the efficacy of chemopreventive intervention, a dermatopathologist indicates the area in a skin shave biopsy considered to be a solar keratotic lesion, and skin of normal appearance, adjacent to solar keratotic lesions. An example is shown in fig 7.

- One of the main advantages of the very large scale digital imagery is the capability to present context in the analysis of a histopathological lesion. The lesion can be examined in context with surrounding normal tissue, an issue that is particularly important in the analysis of focal lesions, such as breast lesions. Figure 8A is a high power field and shows a distended single duct filled with epithelial cells. The changes are difficult to assess and could be benign (two cell types are seen), but in other areas there is a cribriform arrangement suggesting atypical ductal hyperplasia versus ductal carcinoma in situ. Looking at the low power image in fig 8B, there is a background of florid hyperplasia of usual type, and the changes seen in the single duct are easier to identify as benign.

**DISCUSSION**

Our present study shows that there are several technical aspects—such as those related to image acquisition, storage, image compression, networking and line speeds, and computer networks—that can influence the feasibility and accuracy of digital image technology.

For instance, images of large sizes are not practical for transmission via the Internet. Given the capacity and general availability of read/write CD-ROMs, it would have been possible to record the data on a CD and mail it to the consulting pathologist. However, because of the personnel time constraints, it was necessary to have a quick response on the histopathological diagnosis of the scenes so that oil immersion recording of the karyometry data could proceed apace. For this reason, our images were converted from native “ima” format to JPEG to reduce the size of the data file. Alternatively, we could have used “8 bit” colour images, instead of the “24 bit” colour depth.

For example, the image seen in fig 1C, reduced to 8 bit colour depth, has a size of 451 kB, the dimension of pixels being unchanged (TIFF format). This would have restricted the palette to just 256 colours. If the palette provided by the computer system is used, the quality of the image shown on the monitor may be poor. It is possible to define a palette of 256 colours, which is tailored for an individual image. If the variety of colour in the image is limited—as in a haematoxylin and eosin stained section—the result may be visually acceptable, even though the information stored is much reduced. This was demonstrated in a recent paper by Doolittle et al. However, it is known that such a compromise deriving from a restricted number of colours limits the possibility of subsequent image manipulation. For this reason, we decided to use a compression algorithm based on JPEG and to discard the option of “8 bit” colour images.

To receive the images, the reviewing pathologists used the Internet application called FTP. This is a scheme that allows the local computer to connect to the remote host and to transfer files between them. FTP is a set of ground rules (a “protocol”) that allows two computers to exchange files over a network and is supported by hundreds of thousands of machines.

In our present study, this scheme was preferred to an email attachment mainly because, unlike email message/attachment, there is no theoretical upper limit to the amount...
of data to be transmitted during each session. However, FTP is more complex than email because it requires an FTP server running on the recipient computer and FTP software for transmitting files. It also requires the recipient to have a user name and password to allow remote access to the host machine. This prevents unauthorised access and can usually be restricted to a defined directory to prevent users from gaining access to other files on the computer.

Another important factor to be considered in teleconsultation is the type of computer used for image downloading and viewing. This became evident when images were viewed with two other PCs linked to the LAN: a 166 MMX based computer running Windows 95, 32 MB of RAM, with a 17 inch SVGA monitor with millions of colours at 832 by 624 pixels; and a 486 DX/2 based computer running Windows 95, 16 MB of RAM, with a 14 inch VGA monitor, with 256 colours at 640 by 480 resolution. When the images were displayed on the monitor of the former computer, there were no appreciable differences in quality in comparison with those seen with the two Mac computers mentioned in the previous sections. With the last computer, the quality of the images was poor and the display too slow to allow proper evaluation of the images.

In conclusion, the study shows the feasibility of rendering an electronic diagnosis on images downloaded from a remote place. The additional investigations indicated that the same images could be used for further evaluation such as histometry.

**APPENDIX A DETAILS OF COMPUTERS USED BY PATHOLOGISTS TO VIEW IMAGES FOR DIAGNOSTIC PURPOSES**

**Computer 1 (RM)**
The local computer used to download and evaluate the images was a 400 MHz PowerPC G3 (Apple Computers, Cupertino, California, USA) with 104 MB of RAM, a 9 GB hard drive, and running the Mac OS 9.1. The images were shown on a 17 inch colour AppleVision video monitor (Apple Computers) with a spatial resolution of 1152 by 870 pixels and 2 MB of VRAM was used. Each of the three colours, red, green, and blue (RGB), are represented by 8 bits, making it a 24 bit, true colour monitor. This allowed 256 colour values for each plane, or a total of 16.7 million colours.

**Computer 2 (MS)**
A Mac 400 MHz PowerPC G4 (Apple Computers), with 280 MB of RAM, and 10 GB hard disk, and with the Mac OS X operating system was used. The images were shown on a 17 inch colour AppleVision video monitor (Apple Computers), with millions of colours at a spatial resolution of 1024 by 768 pixels, and 2 MB of VRAM.

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Figure 8 (A) A high power field showing a distended single duct filled with epithelial cells. The changes are difficult to assess and could be benign (two cell types are seen), but in areas there is a cribriform arrangement suggesting atypical ductal hyperplasia versus ductal carcinoma in situ. (B) Low power image of same section, which shows a background of florid hyperplasia of usual type, making the changes seen in the single duct easier to identify as benign.
Computer 3 (VDs)

All images received were reviewed at 1024 × 768 spatial resolution and millions of colours (24 bit) on an Intel Pentium II 450 MHz based PC computer (Dell Computers, Austin, Texas, USA) running the Windows 98 operating system (Microsoft, Redmond, Washington, USA) with a 16 MB video card. A Sony Trinitron 200 GS colour monitor (Sony, Tokyo, Japan) was used to display the images.

ACKNOWLEDGEMENTS

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REFERENCES


