Evaluation of virtual microscopy in medical histology teaching

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ABSTRACT

Teaching histology, a major discipline in life science curricula, is based on theoretical didactical strategies as well as on practical training. Traditionally, practical competence is acquired by optical microscopy. Today, information and communication technology solutions are available to study digital microscopic images. Virtual microscopy program was recently introduced at Ghent University. Since little empirical evidence is available about the impact of virtual microscopy (VM) versus optical microscopy (OM) on the acquisition of histological knowledge, the present study was set up in the Faculty of Medicine and Health Sciences. A pretest-posttest and cross-over design was adopted. In the first phase, the experiment yielded two groups in a total population of 199 students, group 1 performing the practical sessions with OM versus group 2 performing the same sessions with VM. In the second phase, research subjects switched conditions. The prior knowledge level of all subjects was assessed with a pretest. Knowledge acquisition was measured with a posttest after each phase (T1 and T2). Analysis of covariance was carried out to study the differential gain in knowledge at T1 and T2, considering the possible differences in prior knowledge at the start of the study. The results point at non-significant differences at T1 and at T2. This supports the assumption that the acquisition of the histology knowledge is independent of the microscopy representation mode (VM versus OM) of the learning material. The conclusion that VM is equivalent to OM offers new directions in view of the ongoing innovation of medical education.
**Key words:** histology education, microscopic anatomy education, medical education, undergraduate medical education, virtual microscopy, optical microscopy, students’ assessment.
INTRODUCTION

Histology is a major discipline within medical and other life science curricula. It can be described as the study of normal tissue morphology. The teaching of histology or microscopic anatomy is partly based on founding theoretical didactical strategies (e.g., lectures), and mainly on practical training that aims at developing experience in identifying tissues (Merk et al., 2010). Actually, the learning objective in general histology is to recognize and to label the microscopic characteristics of all four basic tissue types in the human body (Braun and Kearns, 2008). This is in agreement with the revised taxonomy of Bloom about learning objectives (Krathwohl, 2002). The practice-related competence in histology is traditionally acquired via optical microscopy (with glass slides); hence conventional microscopes have been omnipresent in instructional science laboratories (Coleman, 2009; Paulsen et al., 2010). These days, with the implementation of information and communication technologies (ICT), alternative solutions are available to support the development of the practical competences i.e. digital images of microscopic structures (Heidger et al., 2002; Coleman, 2009). In order to create such digital image files, existing glass slides are scanned at high resolution to be viewed via digital displays. This automatic procedure is called virtual microscopy (VM) (Paulsen et al., 2010). The procedure builds on the integration of conventional optical microscopy (OM) and digital technologies. Conventional optical microscopic systems are still necessary to scan the tissues and record the images, and dedicated software is needed to process them (Bloodgood and Ogilvie, 2006; Braun and Kearns, 2008; Coleman, 2009; Paulsen et al., 2010).

The ubiquitous use of electronic devices is well established in today’s student life and is consistent with the worldwide trend in higher education to adopt online and blended
learning solutions (Ward et al., 2001; Coleman, 2009). The use of digital tools fits perfectly into the current individualization and competence-orientation of higher education programs, e.g., the Five-Star doctor (Boelen, 1996). These developments, together with a decrease in time for basic sciences education (Bergman et al., 2008; Dee, 2009), required many medical schools to redesign their curricula.

At Ghent University (Belgium) a novel curriculum was introduced in 1999 at the Faculty of Medicine and Health Sciences. The overhaul of the curriculum was a direct response to international developments (Drake et al., 2009), and a concrete response to the recommendations offered in 1997 by the External Quality Control Commission (Oosterlinck, 1998). Two key elements were emphasized in the redesigned curriculum. First, at content level, an integrated perspective on medicine was established. Instead of extensively studying separate medicine related disciplines i.e., biology, histology, anatomy, etc., students adopted a multi-disciplinary perspective (Bergman et al., 2008) when studying medical themes (e.g., the cell). Compact learning content is now offered to students as a compilation of several keystones through the associated disciplines. Secondly, at the instructional level, the strategy was directed at more active and collaborative learning (Kumar et al., 2006; Goldberg and Dintzis, 2007; Braun and Kearns, 2008; Collier et al., 2012). The challenge for the present students, also called Millennial Generation students (DiLullo et al, 2011), is to actively extend their knowledge about the learning material outside the classroom and mainly by online resources. As Millennial students are characterized as being agitated and in permanent contact with the outside world through mobile devices, this approach aligns perfectly with their multitasking learning style. Also the time reduction in basic sciences teaching is a change common to curriculum shifts in many higher education institutes.
(Bloodgood and Ogilvie, 2006; Bergman et al., 2008; Dee, 2009; Drake et al., 2009). This affected profoundly the qualitative and quantitative involvement of staff of the Department of Basic Medical Sciences, including the Histology division. Instructional practices were affected quite fundamentally, especially regarding the extent in time to which practical classes (laboratories) were set up. Encouraging home study can be partly considered as a direct consequence of this decline in contact hours. Given these circumstances, the histology staff decided in 2008 to convert from optical (OM) to virtual microscopy (VM).

HISTOLOGY EDUCATION AT GHENT UNIVERSITY: OLD AND NEW FORMAT

Teaching histology or microscopic anatomy obviously is an education assignment of the department of Basic Medical Sciences, since basic knowledge of biological tissues is an essential part of all (bio)medical training programs. Before the curriculum reform in Medicine at Ghent University, only two courses in histology were taught: General Histology and Histology of the Organ Systems. Each of them comprised 45 hours of theory and 30 hours of practice.

Theory was given in plenary lectures by two fulltime faculty members with histological subspecialization. There was no contribution of clinicians or pathologists since it was considered as a basic training in normal human morphology. As a consequence, the histology instructed at Ghent University was purely descriptive: all tissues and organs/organ systems are systematically discussed for the entire audience in a traditional way. Textual and visual illustrations were presented by Powerpoint® slides and explained orally. Questions were posed occasionally by teachers to obtain some student interaction.
The laboratories were performed using the optical microscope with glass slides and a guiding student’s manual. Two teaching assistants are involved to support the students with explanations on request during these microscopy sessions. Usually, giving confirmation about structures students intended to recognize was sufficient. Every session in the microscope laboratory lasted one hour and a half and was devoted to one subject (tissue/organ/organ system). Groups consisted of about fifty students each.

A whole new education strategy is adopted since the curriculum reform in 1999: histology is no longer instructed as a stand-alone course dealing with histology in its entirety, but each tissue/organ is now embedded as a part in several large coherent units (called blocks), in combination with the corresponding cell biology, anatomy and physiology concerning that tissue/organ. This modular and thematic organization of education is in response to progressive changes in medical curricula and promotes horizontal and vertical integration (Bergman et al., 2008; Drake et al., 2009; Amerongen, 2011).

One of the important shifts in the medical curriculum at Ghent University is the serious time reduction in basic sciences practice in favor of more social and clinical practice (Dee, 2009). The sum of all histology lessons in these modules given throughout the academic year is 60 hours of theory (-30%) and 15 hours of practice (-75%!). It is evident that home study is forced in this situation, for which VM is extremely suitable and thus introduced.

Histology remains in essence a descriptive morphological item, and theory is still explained for the full audience. Practical sessions in a multiuse computer space just take one hour and a quarter now and are devoted to more than one subject (tissue/organ/organ system). Groups are now enlarged to one hundred students at a time, supervised by only one teaching assistant.
Written examination on different histology topics takes place at the end of the semester. Several structures have to be recognized on printed static photographs. Students pass this summative practical exam when at least half of the corresponding multiple-choice questions is correctly answered. If desired, personal feedback is given in a one-to-one conversation with the teacher in case of unsatisfactory marks.

DIGITIZATION OF GLASS SLIDES AT GHENT UNIVERSITY

Virtual Microscopy offers the experience of handling a real microscope by using digitized slides, also called virtual slides (Bloodgood and Ogilvie, 2006; Dee, 2009). To create virtual slides, specific equipment is needed, consisting of a microscope with a motorized stage, different lenses, a digital camera, software that manages stitching, compressing and viewing (viewer program) and a dedicated large capacity storage system (server) (Coleman, 2009; Paulsen et al., 2010). In 2006, the Ghent University Histology Department started to scan glass slides in view of developing the experimental setting described below. Thanks to additional grants from the Faculty, the advanced virtual slide system, called dotSlide® (Olympus Europa Holding GmbH, Hamburg, Germany), was purchased in 2008. Currently, all glass slides used for instructional purposes (comprising all tracts of the human body) have been scanned and converted into digital image files. As a result, VM could be fully implemented in all morphology related courses. The virtual slides are stored on a terminal server in the ICT Department of Ghent University and are accessible via the online application platform at https://athena.ugent.be.
VIRTUAL MICROSCOPY REPLACING OPTICAL MICROSCOPY: BENEFITS OR THREATS?

In the former sections, a rather limited rationale was presented to discuss the shift from VM to OM. In the literature, extensive lists about the potential advantages of VM can be found.

We cluster the major benefits and discussion points as follows:

- Thanks to the World Wide Web, access to virtual microscopy is independent of time (opening hours) and place (institute) (Harris et al., 2001; Leong and McGee, 2001; Blake et al., 2003; Krippendorf and Lough, 2005; Coleman, 2009; Merk et al., 2010; Paulsen et al., 2010). So there is no longer need for formal localized laboratory sessions and home study becomes possible (Harris et al., 2001; Heidger et al., 2002, Blake et al., 2003; Merk et al., 2010), either autonomously or under supervision from a tutor (whether or not real-time).

The integrated usage of the Internet also allows for interaction and communication (Heidger et al., 2002; Paulsen et al., 2010), within and outside the classroom setting.

- The number of users that can examine specimens simultaneously is limitless. This fact is important in a variety of educational contexts: collaborative learning/group work/team-based learning (Harris et al., 2001; Heidger et al., 2002; Blake et al., 2003; Kumar et al., 2006; Goldberg and Dintzis, 2007; Braun and Kearns, 2008; Collier et al., 2012); remote education/distance learning/teleclassing (Krippendorf and Lough, 2005; Pinder et al., 2008; Coleman, 2009; Merk et al., 2010; Paulsen et al., 2010) as well as in a clinical setting (second opinion in pathology, quality control, scientific meetings: Harris et al., 2001; Leong and McGee, 2001; Krippendorf and Lough, 2005; Coleman, 2009; Lundin et al., 2009, Paulsen et al., 2010).

- The digital study of histological slides on a monitor offers new possibilities for specimen viewing and handling: different or special stains can be shown in parallel or in overlay on the
same screen (Paulsen et al., 2010), an entire slide can be viewed, scanning in Z-stack modus is the base for 3-D reconstruction of images (Dee, 2009; Paulsen et al., 2010), ... The Z-stack modus is a multiple focus layer image acquisition option of the scanning system which permits to view the obtained image in several well focused planes. All these modalities are not possible when using traditional optical microscopy.

- Thanks to the software, an overview image remains present on the screen while zooming in on the slide. This facilitates orientation and navigation (Blake et al., 2003; Kumar et al., 2006; Paulsen et al., 2010).

- Markings and textual explanations (annotations) can be superimposed (Dee, 2009; Merk et al., 2010; Paulsen et al., 2010). This labeling of specimens enhances the (inter)active study and feedback possibilities to students.

- Additional textual or other visual information about the slide can be added to the database e.g., gross anatomy images, electron microscopic pictures, relevant clinical examples, digital radiographic data (Harris et al., 2001; Paulsen et al., 2010). Even a complete student laboratory manual can accompany the slides (Heidger et al., 2002; Blake et al., 2003; Coleman, 2009), providing a link between annotations on the image and microscopic terminology in the text. This combination of information stimulates horizontal integration (Harris et al., 2001; Kumar et al., 2006) between basic medical sciences (histology, anatomy, physiology) and between biomedical sciences and clinical disciplines. At the same time, it presents a pathway to ‘e-learning’.

- Computer-assisted learning (CAL) (Ward et al., 2001), as described just above, also allows for vertical integration, i.e. the integration across curriculum years. This results e.g., in students of year 1 (histology) and year 2 (histopathology) studying materials together
This peer teaching encourages sharing perspectives, discussion and collaborative learning / group work / team-based learning (Blake et al., 2003; Kumar et al. 2006; McBride and Prayson, 2008; Amerongen, 2011; Shaw and Friedman, 2012).

- Alternative learning and evaluation modes become possible. Teaching and testing evolve into a problem-based (PBL) approach by e.g., incorporating virtual slides into case studies (Kumar et al., 2006; Coleman, 2009; Paulsen et al., 2010). A linkage between instruction and evaluation offers advantages such as automated correction, uniformity in slide presentation, accessibility, re-usable sets of test-related images (archives / collections) (Harris et al., 2001; Heidger et al., 2002; Blake et al., 2003; Coleman, 2009; Merk et al., 2010; Paulsen et al., 2010).

- Finally, the electronic way of visualizing histological slides diminishes the costs for maintaining a microscopy laboratory (Krippendorf and Lough, 2005; Paulsen et al., 2010). There is no need for individual microscopes nor a set of glass slides for every student (Coleman, 2009). As a consequence, considerable long term savings in teaching budgets appear (Coleman, 2009). At the short term, investment is needed in student computer facilities (Dee, 2009), in suitable hardware and software to scan high resolution slides in the morphological laboratory unit (Goldberg and Dintzis, 2007; Coleman, 2009), in the procurement of special viewer programs, appropriate monitors, fast transmission network (Paulsen et al., 2010) and adequate server space.

In summary, virtual microscopy as a modern ICT tool has the potential to affect in a profound way teaching and learning environments in medical education (Ward et al., 2001; Valcke and De Wever, 2006).
IMPACT OF VIRTUAL MICROSCOPY ON LEARNING PERFORMANCE

When it comes to the expected impact on learning performance, the research evidence to ground the theory-based rationale in the VM-literature remains scarce. The learning benefits presented by VM can be regarded from a cognitive psychological perspective stressing the need for multiple representations to develop sufficiently rich schema during knowledge construction (Feltovich et al., 1989). The emphasis on the visual nature of the representations is also consistent with schema theorists who stress ‘visual learning’ and indicate that presenting multiple representations will foster differentiation and interpretation (Couch et al., 1994). The different viewing possibilities with VM foster the decoding processes in working memory to identify and recognize relevant visual information and to classify this information. The fact that VM allows enrichment of the visual presentation with additional knowledge elements is in line with the benefits stated by the Cognitive Theory of Multimedia Learning (Mayer, 2001) which stresses the advantages of exploiting multiple sensory channels to foster the organization and integration of cognitive schema. In contrast, OM also presents benefits. Though the ‘visual learning’ dimension is more limited and OM does not allow for a large extent of flexibility, OM nevertheless presents the possibility to be actively engaged in the interaction with a physical microscope. This can be referred to as the ‘activation’ principle. Several studies stress that learners often remain passive when faced with multimedia representations (Bodemer et al., 2004). In contrast, instructional interventions that promote the active creation of personal multimedia representations are expected to counter this. Marzano et al. (2001) present a meta-analysis of studies that tested the active construction of non-linguistic representations (NLR) and report effect sizes varying from 0.5 to 1.3 on learning performance. But these hypotheses
have not been tested in the context of microscopy. Other authors stress that OM presents the benefit of developing microscope manipulation skills in medical students; considering the fact that the microscope remains an important ‘tool’ (Krippendorf and Lough, 2005; Goldberg and Dintzis, 2007; Braun and Kearns, 2008; Coleman, 2009; Paulsen et al., 2010). In the present study we do not focus on these manipulation skills, but centre on the impact of VM on histology related knowledge acquisition. Nevertheless, due to the cross-over nature of our research design, we are able to test the potential ‘preparatory’ impact of OM related knowledge acquisition on resulting VM related learning opportunities. We can compare whether students, starting with OM and continuing with VM, outperform students who immediately started with VM and switched to OM.

As stated earlier, in sharp contrast to the literature about the potential of VM, there is a lack of empirical evidence about the impact of VM on learning performance. Especially evaluative data are lacking that explore the impact of switching from OM to VM in histology teaching (Scoville and Buskirk, 2007). The need for systematic evaluation studies in relation to VM is repeatedly stated in the literature (Paulsen et al., 2010). The latter is especially put forward in the context of curriculum reforms (Krippendorf and Lough, 2005; Bloodgood and Ogilvie, 2006). As a result, histologists and pathologists remain skeptical as to the introduction of VM. They criticize the loss in ‘hands-on’ microscopy skills of students (Coleman, 2009; Paulsen et al., 2010) and expect a decline in learning performance. Authors stress that VM should at least present the same educational value as OM (Heidger et al., 2002, Paulsen et al., 2010). This brings us to the focus of the present study about the differential impact of studying via VM or OM on learning performance of students enrolled in medical curricula. The study was set up in partnership with the Faculty of Psychology and Educational Sciences.
RESEARCH DESIGN

An experimental pretest-posttest and cross-over design was adopted to study the impact of the implementation of VM versus OM on the acquisition of histology knowledge.

Research hypothesis

Building on the theoretical base as to the expected impact of VM and OM, the following hypothesis is put forward: Studying histology via VM will result in at least equal learning results as studying via OM.

Participants

The study was based on data acquisition during a time span of two academic years (2006-2007 and 2007-2008). This allowed involvement of three different student populations from the Faculty of Medicine and Health Sciences at the University of Ghent in which the same research design was applied: all 1st bachelor year students in Biomedical Sciences 2006-2007 (n = 172), all 1st bachelor year students in Biomedical Sciences 2007-2008 (n = 202), and all 1st bachelor year students in Logopaedic and Audiological Sciences 2007-2008 (n = 104).

Exactly the same practical histology course is a formal part in the curricula of these three study programs. Because of this, the sum of the three mentioned numbers of students (n = 172, n = 202 and n = 104) can be considered as the starting size of the overall research population under study (n = 478). Of this initial total population, 121 students were repeaters or non-debutants with histology in their previous curriculum. Considering their prior knowledge about histology, data from these students – though they participated in the study – were excluded from the analysis. This resulted in 357 freshman participants being
involved in the present study, belonging to the three described subpopulations. Since the research design and procedure required participants to be involved in four practical sessions, organized during four successive weeks in one semester, a number of students did not participate in all sessions, resulting in missing values in the overall data set. After exclusion of students who dropped out during the experiment, a dataset of 199 valid cases out of 357 starting students could be included in the analysis, among them 170 female and 29 male students with a mean age of 19.24 (SD 0.82) years. Figure 1 summarizes the breakdown in the student participation in the research procedure. Non-participation or incomplete participation was related to individual circumstances, and especially due to time scheduling problems. It was not related to specific background characteristics.

**Research procedure**

Informed consent was obtained from all students at the start of the research study. The ethical committee of the Faculty of Medicine and Health Sciences at Ghent University approved the study. The investigation consisted of three iterations of the same research design, each time involving one of the three research subpopulations (see above). In each experiment, the students were assigned randomly to one of the two research conditions (optical or virtual microscopy), simply based on the alphabetical order in the list of names. Students participated in four consecutive practical histology sessions as follows: they completed two times a short series of two sessions in the same condition. Considering the cross-over nature of the research design, participants switched research conditions after the first two sessions. This resulted in two learning sequences: an optical-virtual microscopy sequence and a virtual-optical microscopy sequence. This is depicted in a graphical way in
Figure 2. Students in group 1 (n = 95) started in the OM condition and continued in the VM condition. Students in group 2 (n = 104) started in the VM condition and continued in the OM condition. The cross-over design was applied first of all in consideration of ethical conditions to guarantee that students received equal learning opportunities. But as stated in the theoretical introduction, the cross-over design also helps to study sequencing effects of e.g., starting with OM and continuing with VM. Therefore, next to a pretest at T0, test results after session 1 and 2 were summed to obtain a T1 posttest score, and at T2 the test results after session 3 and 4 were summed to obtain a T2 posttest score.

Histology learning environment during the experiments

The learning content subject in the consecutive practical sessions was the same in each research condition: session 1. epithelial and glandular tissue; session 2. connective tissue and cartilage; session 3. bone and muscle tissue and session 4. nervous tissue and blood.

Each laboratory session lasted two hours and took place in a classroom setting. Students in the two opposite research groups handled identical content on the same day in order to limit interfering factors. Sessions were constantly monitored by one and the same academic staff member who followed a standardized protocol to give instructions as to the hands-on student activities. More specifically, a 20-min introductory lecture with Powerpoint® slides was given in the two groups. Students were not considered to prepare the sessions in advance. Neither they were allowed to use additional resources such as atlases or textbooks during the practical nor the test sessions. Instead, they could only consult the staff’s practical notes containing a short description of all microscopic structures in each slide to view. The exposure to the optical or virtual slides occurred only during histology laboratory
contact hours, so that students were keeping an open mind about the slides. Thanks to on-off interventions by the ICT Department staff at regular intervals, access to the virtual slides was absolutely restricted to the duration of the practical classes themselves. Outside of scheduled class time, no student could view the virtual images. During laboratory time, only the slide viewer software OlyVIA® (Olympus Europa Holding GmbH, Hamburg, Germany) was within reach via the online application platform of Ghent University whereas other websites were locked.

Obviously, the same principle applied at the optical side. Since all boxes with glass slides for educational purposes are kept under lock and key at the University, no student could use them for home study outside the provided contact time. Besides, all virtual slides made available in this investigation were created outgoing from the corresponding glass slides via the automatic scanning procedure as described earlier. So during the period of observations, both the OM group and the VM group had the opportunity to view the exact same slides with the exact same microscopic capabilities in each instruction system.

**Research variables and research instruments**

The central dependent variable consisted of the measurement of knowledge acquisition in relation to the histology themes covered in each practical session. Each session posttest consisted of 4 or 5 test items (printed images on a sheet with 6 to 10 related brief questions) requiring learners to visually recognize tissues on the base of static photographs. An open ended question format was applied to guarantee the active production of the related knowledge elements. In each posttest, items focused on the tissues studied during the particular session. Each test item, and the related tissue high-resolution photo was assessed
by a team of three histology specialists to guarantee content validity. The correction of student responses to test items was based on a correction key. This time consuming correction procedure guaranteed a systematic and reliable correction and scoring of student responses. Considering the differences in the number of test items in the different posttests, absolute scores were all converted into relative scores to 10. This standardization procedure allowed comparative analyses. The students involved were not being graded on the basis of their participation nor received any kind of feedback to their test results, because of the anonymity principle implied in the guidelines of the approving ethical committee. To assess the prior knowledge about histological tissues, an unannounced pretest was presented to the students prior to the experiment. Before any theoretical or practical histology lecture, the plenary class had to complete the pretest form individually. The pretest was presented via static photographs (7 items) accompanied with short questions which covered the full range of all histological tissues that would be studied during the subsequent four practical sessions. Pretest scores were also normalized to 10 for analytical reasons. Random division of student subjects in two research groups occurred afterwards on an alphabetical base. The pretest and all the posttests were set up in an analogous way. Both were administered on paper not to favor one or another microscope research condition. All the questions were open-ended and served as a guideline for the students as well as an objective assessment tool for the teaching staff. In the pretest, questions were rather of a general nature, such as “Which human tissue do you recognize on this photograph?”, while questions in the different posttests were specifically directed to the tissue studied in the practical session concerned e.g., “Which type of epithelium do you recognize?”. 
Moreover, test questions were not conceived as in a classical histology examination, but relied directly on the visual part of the working memory of humans. In other words, students were definitely not given any time period to assimilate the learning material outside laboratory contact time.

**Statistical analysis**

All statistical analyses were performed using SPSS Statistics 15.0 for Windows® (SPSS Inc., Chicago, IL). To compare the posttest scores of students in both research conditions, and considering the potential differences in prior knowledge at the start of the experiment, analysis of covariance (ANCOVA) was applied. The analysis was carried out twice: (1) on the combined posttest scores after the first two sessions (T1) and (2) on the combined posttest scores obtained after the last two sessions (T2). The following assumptions were tested: (a) independence of observations; (b) normal distribution of the dependent variables within each group (Kolmogorov-Smirnov test); (c) homogeneity or equality of variances (Levene’s test); (d) the linear relationship between the covariate and the dependent variable and (e) the homogeneity of regression slopes. In case assumptions were violated, alternative analysis approaches were adopted. A significance level of \( p < 0.01 \) was put forward.
RESULTS

Descriptive results

Table 1 gives an overview of the descriptive results in relation to the different measures investigated in the study. The results in the table suggest a differential impact of the microscopy mode on learning performance at T1 and T2.

Differential impact of the research conditions on knowledge acquisition

In a first analysis step, we study potential differences already present at the start, despite the fact that students were assigned at random to the research conditions. The results of an independent samples t-test point at non-significant differences in T0 pretest scores between the two groups at the start of the experiment \( (p = 0.184) \), confirming that random assignment by alphabetical order is a valid method to select groups. Secondly, we focus on the potential differences observed at T1 between the test results of students in both research conditions and considering the differences in the pretest results (T0). The pretest score at T0 is thus considered as a covariate in the ANCOVA to correct for the existing differences between individual students at the start of the study.

At T1, the results of the analysis of covariance point at non-significant differences in knowledge acquisition between the two research conditions: \( F(1,196) = 0.465, p = 0.496 \) (significance level \( p < 0.05 \)). A Cohen’s d effect size value of 0.125 (< 0.3) implies that there is no effect of microscopy mode on learning performance.

Thirdly, the analysis focuses on the potential differences observed at the end of the experiment (T2). As stated earlier, these results could help to point at sequencing effects of studying first OM or VM on results obtained after studying in a next phase via a different...
microscopy mode. Therefore, in the analysis of covariance, we consider both the pretest results and the results obtained at T1 as covariates.

At T2, the analysis results point again at non-significant differences between the two ways of microscopy learning: F (1,195) = 2.657, p = 0.105 (significance level p < 0.05). A Cohen’s d of 0.253 (< 0.3) validates this finding. The ANCOVA numerical data are represented in Table 2.

In short, the lack of a clear impact on learning performance evidences the equivalence of VM and OM conditions. Moreover, these results suggest sufficient proof to accept that at the end of the total experimental period, students who studied with OM during the first research phase don’t attain significantly higher scores than those who studied with VM during that first phase. In other words, students who started with OM don’t outperform students who started with VM. Thus both learning sequences OM-VM versus VM-OM end in the same final level of histology knowledge.

The analysis results are depicted graphically in Figure 3. Next to the pretest results, adjusted average means are represented at T1 and T2.

In summary, neither the nature of the medium nor the order of use seems to be important to transfer adequately the histology learning material.
DISCUSSION

Today, virtual microscopy (VM) is finding more and more acceptance as a promising instructional tool in morphological sciences. Apparently in the USA, but also in several European countries (Dee, 2009; Lundin et al., 2009; Merk et al., 2010; Paulsen et al., 2010; Helle et al., 2011) it is likely to replace steadily the long-established optical microscopy (OM) in (bio)medical education. The above inquiry executed at Ghent University in Belgium aimed to investigate whether modern virtual microscopy can compete with traditional optical microscopy regarding the acquisition of histology recognition skills. The reason for converting our instructional practices from OM to VM was the overall decline in laboratory contact hours (Bloodgood and Ogilvie, 2006; Bergman et al., 2008; Dee, 2009; Drake et al., 2009), as confirmed with the introduction of a contemporary curriculum.

The obtained results reveal no significant differences in learning performance between OM and VM instruction mode. At first sight, these findings confirm those of earlier studies (Blake et al., 2003; Krippendorf and Lough, 2005; Scoville and Buskirk, 2007; Braun and Kearns, 2008; Dee, 2009). Indeed, the number of papers concerning comparisons between OM and VM in microscopic anatomy education, has increased rapidly the last few years (Dee, 2009). However, many of them deal with observational reports on subjective data about the daily use of VM versus OM by students (Goldberg and Dintzis, 2007; Braun and Kearns, 2008; Husmann et al., 2009; Merk et al., 2010; Helle et al., 2011). Our comparative study conversely presents an attempt to fill the still existing gap in available empirical evidence about the use of virtual versus optical microscopy in medical education (Paulsen et al., 2010). Even though experimental studies with objective numerical outcome are published on this issue, they always compare single formal test results obtained after a once-only
regular examination either with OM or with VM (Goldberg and Dintzis, 2007; Scoville and Buskirk, 2007; Braun and Kearns, 2008). Our experimental setting is unique because of several reasons. Results are not based on comparisons of single examination scores between student cohorts, but are obtained after several measurements (i.e. after each practical session in a series of four) within the same student cohort. Little similar research is done. In addition, the measuring didn’t rely nor on VM neither on OM tissue recognition, as a traditional examination does. In our design, the testing itself is strictly considered not to be favoring one or the other condition, and is therefore taken by photographs on paper. Moreover, test questions relied directly on the visual part of the working memory of humans. So students were definitely not given any time to assimilate the learning material outside laboratory contact time, nor a consolidating period of sleep, which is surely the case with classical histology examinations mentioned in the VM-literature. As just mentioned, our experiment reveals no significant differences between post practicum scores after following either optical or virtual microscopy sessions in histological practice. The results imply that the histological knowledge acquired during this sessions seems to be independent of the representation modus of the learning material. VM can be considered as being equivalent to OM as a teaching tool in histology, based on microscopic visualization (Heidger et al., 2002; Paulsen et al., 2010). In other words, although it didn’t yield higher scores, VM is at least as effective as OM, even when not all its potentials (see Benefits) were exploited in this setting in order to maintain equal research conditions. Moreover, the learning sequence seems to be of no importance regarding the learning outcome. A possible explanation is exactly the rough use of VM in this study, without accessory advantages against OM. As a consequence, VM should not be seen just as a
compensation tool for the ‘loss’ of OM, but can be considered as a full alternative for OM, even to start with a histology instruction series. Even more importantly, VM can involve an added value when used at its full potential (see Benefits).

A possible limitation has to be stressed in relation to the present study. Our data set seems to be rather limited, not in terms of participants, but in terms of timespan and number of questions included in the posttests. The time period during which the intervention (i.e. four practical sessions) could take place was quite short, namely one semester, because of the university timetable. Also the learning content (four basic tissue types in General Histology) is a limiting factor because it is suitable for only a restricted number of practical sessions (four times two hours). Each posttest taken immediately after the session, contains 4 or 5 photographs with 6 to 10 associated questions, which is a sufficient number to cover all the learning material of the subject concerned. The sum of all questions to be answered by each student during the whole intervention period is 30, which is still a statistically acceptable number. So despite of this minor limitation, we can state that the limited time frame and the restricted number of test items seem sufficient to adequately interpret our results.

Though our findings are promising, they do not suggest we should replace OM completely. Indeed, OM offers some specific advantages due to the more authentic handling procedure for viewing glass slides. For instance, users of OM are always able to focus up and down in a section, whereas users of VM can only do so after a special scanning procedure called Z-stack modus, which is not a routine acquisition option (Goldberg and Dintzis, 2007; Dee, 2009). Thin tissue sections in the histology laboratory are produced through cutting up a whole paraffin embedded tissue part by means of a microtome. So students are working with slightly different serial sections, which can be an incentive for mutual discussions
between neighbouring students using OM. Finally, one can assume that the search for structures is more explicit with OM than with VM because of the lack in OM of a permanent overview image in a software navigator window on the screen. Strictly speaking, these features of OM are not necessary to achieve the learning objective to recognize basic tissue types. Moreover, the focus in our study was not on technical competences that might still be critical in view of attaining microscopy related skills (Krippendorf and Lough, 2005; Coleman, 2009; Pratt, 2009; Paulsen et al., 2010; Collier et al., 2012).

Establishing the principle of ‘blended learning’ presents a possible answer to the dilemma stated above. VM is adopted to be implemented in a distributed and flexible learning environment, but OM keeps being adopted in view of guaranteeing the development of related competences (Goldberg and Dintzis, 2007; Braun and Kearns, 2008; Pratt, 2009). For reasons of certainty, most institutions have adopted such a mixture of OM and VM for histology laboratory instruction (Bloodgood and Ogilvie, 2006; Braun and Kearns, 2008; Dee, 2009, Merk et al., 2010), so has the University of Ghent. Hence, optical microscopes are still available for our students in Biomedical sciences, who will perform executive tasks in their future laboratory-oriented profession. In addition, ‘blended learning’ is in conformity with the earlier mentioned fact that multiple representations of visual material will aid learning (Couch et al., 1994).

Because teachers of our department could not escape the subjective impression that learners with the basic version of VM show a rather passive attitude towards studying histological tissues on the screen (Bodemer et al., 2004; Collier et al., 2012), further research is needed to investigate the active processing in students working on the base of VM images. Related competences have to go beyond the recognition and knowledge about histological
tissues (Krippendorf and Lough, 2005; Bloodgood and Ogilvie, 2006; Coleman, 2009). Students should be able to work with the VM identified structures and to apply them subsequently in other - slightly different - tissue settings. In order to stimulate these competences, the Histology Department at Ghent University will design, implement and evaluate ‘activation’ measures during practicals; e.g., online guiding and self-assessment questions (Dee, 2009; Collier et al., 2012) during and after VM practice, and the integrated use of VM in case-based (CBL) and problem-based learning (PBL) (McBride and Prayson, 2008; Drake et al., 2009), which clearly represent two aspects of the mentioned added value of VM.

In this multimedia age, it is no accident that one of the educational themes in the large-scale strategic plan of Ghent University, called Growth Plan (2012-2016), is defined as ‘active learning’. VM perfectly lends itself to do so (Goldberg and Dintzis, 2007; Amerongen, 2011; Collier et al., 2012) on condition that it will be fully exploited in the future.

Finally, despite the pros and cons of both VM and OM, two things remain of greatest value for students in morphological sciences: having an excellent original slide (Coleman, 2009) and above all a passionate teacher (Heidger et al., 2002; Bloodgood and Ogilvie, 2006; Scoville and Buskirk, 2007; Braun and Kearns, 2008).
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LITERATURE CITED


FIGURE LEGENDS

Figure 1: Flow chart with drop out of participants in the study.

Figure 2: Nature of the cross-over design and timing of testing.

Figure 3: Graphical representation of the impact of different conditions on T1 posttest score and T2 posttest score, considering differences in the T0 pretest results.