Competence-based education in the example of a biochemical practical course with clinically relevant content for undergraduate medical students in the fourth semester of the preclinical section

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Categories: Educational Strategies, Learning Outcomes/Competency, Teaching and Learning, Undergraduate/Graduate

Received: 10/08/2018
Published: 14/08/2018

Abstract

Focus in traditional practical courses has always been on imparting technical knowledge. However, significance and application often are unclear for students, leading to low motivation. Thus, a practical course that is clinically relevant to students in the 4th semester was set up in biochemistry so that: a) a clear link to the clinical work can be identified, b) the roles of the physician, as set out in the German National Competence-based Learning Objectives Catalogue for Undergraduate Medical Education (NKLM), are covered in the best possible manner, and c) the scientific basis has a high importance.

The newly established course covers key points of research work in 4 days, beginning with a fictitious case study. Samples of (artificial) urine and blood are used to diagnose sickle cell anemia and malaria, and their molecular basis is analysed using bioinformatic tools. Students learn how to research and discuss academic publications. Working in groups, they establish a concept using gene therapy to explore how a higher expression of fetal hemoglobin in erythroblasts could be induced using RNA interference to attenuate progression of the disease. Instead of recording the findings in a traditional manner, the students work together in groups to produce a manuscript of their results, which is critically examined to assess the suitability for being published.

The semester cohort was randomly allocated to the newly established (n=79) and the traditional (n=57) practical course. Both courses were comparatively evaluated. The newly designed course received an overall assessment of 1.4, which was significantly better than the traditional course which received an overall assessment of 2.9.

During the next years, nearly all subjects of the preclinical section will be challenged to find ways of combining NLKM requirements and technical knowledge. The concept presented here may serve as a model to be readily adapted to other fields.
Keywords: biochemistry; practical course; NKLM; competence-based medical education; curriculum development

1. Background

From summer 2011 to winter 2015/16, the Department of Pharmacy and Biochemistry of the Eberhard-Karls-University of Tübingen has offered a practical course in the fourth semester for medical students, which covered four course days with the content of each not linked to that of the others. Although experiments for the modules liver/kidney/urine, molecular diagnostics, blood and bioinformatics were performed, the clinical relevance was implicit, and the course days were not integrated into the overall context. Despite the high level of commitment by the lecturers, the students gave the biochemistry practical course only a moderate rating (Figure 1). This suggests that the organizational structure and format of the events did not (or no longer) meet the expectations of the participants. It was in fact very clear that only a few students developed sufficient interest in the experimental details. Correspondingly, the tasks often led to incomplete results, which meant that their educational use was questionable and barely recognizable for the students. However, solid basic knowledge of biochemical and molecular biological interactions is an elementary component of medicine and is essential for diagnosis and therapy. Thus, the dilemma was that the access to an experimental science should logically take place through the design of adequate tests and independent experimentation, but this was only unsatisfactorily communicated in the framework of the existing practical training model. As a result, the relevance of the employed laboratory techniques was often underestimated. The students found it increasingly difficult to recognize the application areas and to establish points linking to the pursued activity of a practicing physician. Through the establishment of modular study programs (Thiel, 2017), in recent years a further development of the medical degree was initiated, during which the traditional subject-oriented training should be replaced by training oriented to the roles of the physician and their expertise (NKLM, 2015). The more recent modular programs primarily emphasize strengthening the ability to perform independent research. By gaining scientific expertise, the prospective physicians are put in the position where they can make evidence-based decisions using the respective current state of medical science. The German science council explicitly recommends placing even greater emphasis on scientific expertise in the corresponding specific lectures (Wissenschaftsrat, 2014).
Figure 1: Overall assessment of both practical courses "old" and "new" during the last six years.

2. Methodology

2.1 Positioning of the event in the curriculum

In the second semester of human medicine, the students begin with the introductory biochemistry lecture, which covers the topics cell biology (structure and function of membranes, proteins, DNA and carbohydrates) and energy metabolism. Additionally, in the second semester there is a basic practical course to introduce the students to practical work in a biochemistry laboratory. This takes place in the form of experiments, which are grouped according to the biologically-relevant basic components. The main lecture in the fourth semester expands the topic to cover the areas hormones, proteolysis (digestion, intracellular protein breakdown, blood coagulation cascades), immunology, and intermediate metabolism. The practical course presented here runs parallel to this in the fourth semester. Full and successful participation in both lectures and the two practical courses leads to the student receiving the certificate of achievement “Praktikum Biochemie und Molekularbiologie”.

2.2 Educational concept

High motivation and commitment are generally achieved when the students can see the benefit of the event for the later professional life. For this reason, the emphasis of the practical course was placed on:

a. demonstrating a clear clinical function. This was achieved through the diagnosis of an illness using artificial “patient material” (i.e. blood or urine which had a similar appearance to that of real patient samples and were suitable for the experiments) and the design of a concept for the development of a previously unavailable gene therapy.

b. offering a coherent context so that the students were able to recognize the aim of their work. After examination, analysis and molecular biological research on a fictitious patient, the students compiled a manuscript as teamwork (each small group had a sub-point), which had a similar appearance to that of a publication.

c. having no classical hierarchy (lecturer-student), but instead the students were motivated to perform independent work. Individual topic areas were worked on by small groups and presented to the other participants. In this way, the students naturally carried a high degree of responsibility towards the whole group. The lecturer was ideally only there as a moderator, however, was always available for questions and discussions.

d. it being carried out by one lecturer per practical group, who supervised the participants on all four practical days. In this way, the individual students could be supported better and receive sound feedback.

2.3 Recruitment method and evaluation

The biochemical practical course is mandatory in the curriculum at the University of Tübingen. Therefore, inclusion criterion for this study was being an enrolled medical student in the fourth semester of human medicine in Tübingen. The practical course took place in the seminar and practical rooms of the Interfaculty Institute of Biochemistry (Tübingen, Germany).

After completion of the event, participants are routinely invited via email to fill out an online questionnaire. This questionnaire is provided by the medical faculty using the evaluation software Tuevalon (Ostrakon Software
GmbH). The traditional practical course was evaluated by 742 participants during summer semester 2011 to winter semester 2015/16, the new course was evaluated by 229 participants from winter 2016/17 to winter 2017/18. In summer 2016, students in the fourth semester of human medicine were randomly allocated to the traditional (n=57) and the newly established (n=79) practical course. After completion of the event, printed questionnaires were handed out to evaluate specific aspects of both practical courses. Participation was anonymous and voluntary. Regarding response rate and acquisition time, this data can be considered representative of a larger population.

2.4 Course of the event

2.4.1 Practical day “Patient presentation and laboratory diagnostics” (Day 1)

The first day of the practical (Figure 2) started with an introduction of a fictitious 30-year-old patient of African descent (180cm, 75kg), who complained of reoccurring headaches and aching limbs after returning from a trip to the Congo. Body temperature was 39°C. In the case history, he also indicated that he previously suffered from breathlessness and reduced capacity to cope with stress. After physical exercise, he observed lightly foaming urine on several occasions. The patient said he was a low smoker and only consumed alcohol in moderation.
| Seminar | Experimental procedure | Debriefing |
|---------|------------------------|------------|
| 2.5 h   | 2.5 h                  | 1 h        |

**Presentation of a fictitious patient**

- 30-year-old male, normal body mass index, African origin, recurring flu-like symptoms after trip to Congo, fever, breathlessness, foaming urine, little nicotine/alcohol

**Elaboration of possible differential diagnoses & molecular markers**

- Lecture-moderated seminar with creation of an advance organizer at the blackboard. (Fig. 3)

**Group work**

- Lactate dehydrogenase (Fritz, 1985), LDH subunits (Adams, 1976), measurement of activity
- Amino transferases, optical test of coupled enzymatic systems (Karmen, 1955; Huang, 2006)
- Use of hemocytometer (Marienfeld, 2017), Hb-measurement (Zander, 1984)
- Glucose measurement (Keller, 1965), meaning of HbA1c (Pundir, 2014)
- Hb-electrophoresis (Basset, 1978), isoelectric focusing (Wikipedia IEF)
- DNA isolation from blood (Butler, 2012; QIAGEN, 2017)
- Plasmodia detection in blood smear, malaria and plasmodia lifecycle (Bray, 1982; Wikipedia Malaria)
- HIV test, ELISA (Fearon, 2005; Gauthier, 1989)
- Creatinine- (Narayanan, 1980), protein- (Goldring, 2012)
- Determination in urine

**Performance of assays in small groups**

- Approx. 3 persons per assay.

- Eight work benches with a full set of pipettes/chemicals/etc. are available. Each bench corresponds to one distinct assay.

- Each small group can choose an assay according to their interest.

**Boundary conditions:**

- Depending on the duration of an assay, several assays should be completed.
- All assays should be covered by the whole group (i.e., 8-10 small groups).

- Detection of plasmodia
  - A pre-fixed/stained blood smear slide with P. vivax (Johannes Lieder GmbH) is used.

- Erythrocyte count of anemic blood
  - Neubauer-counting chamber
  - A pre-diluted blood sample is handed out to give diagnosis anemia (i.e., a cell count of 500000 per μl)

- Creatinine and protein determination
  - Artificial urine: measurement of extinction after reaction with picric acid (normal value), Bradford-assay (diagnosis proteinuria)

- DNA-isolation from whole blood
  - DNA concentration and purity are measured; sample is said to be used for HBB-sequencing; sequence will be analysed during Day 2.

- Hemoglobin-electrophoresis
  - Isolation of Hb from blood which contains HbA and HbS (Sigma-Aldrich) by hypotonic lysis and chloroform delipidation, agarose-electrophoresis, diagnosis sickle cell anemia

- Blood glucose determination
  - Extinction measurement of NADPH after glucose-6-phosphate dehydrogenase reaction, normal glucose value is found

- Lactate dehydrogenase determination
  - Extinction measurement of NAD. LDH (Sigma-Aldrich) had previously been added to give diagnosis hemolytic anemia

- HIV-ELISA
  - Serum sample had been prepared to give negative result

**Presentation of primary data in small groups and discussion of the results**

- Lecture-moderated seminar: insertion of results into the advance organizer from the initial seminar, Fig. 3, and giving a diagnosis.

**Presentation of group work**

(Aim: Comprehension of the fundamental assay principles, max. 5 minutes/group)

- Available media: blackboard, interactive whiteboard (tablet PC with WLAN connection to beamer)

**Logging (primary data/calculations)**

- Directly into designated fields of the course notes.
  - (Free space for notes / printed millimeter paper)
Figure 2: Organization of Day 1: “Patient presentation and laboratory diagnostics”

The symptoms, suitable laboratory diagnostic procedures and potential diagnoses were discussed in an around 20-minute seminar using the Think-Pair-Share method. The lecturer developed an Advanced organizer on the board from the students’ contributions to the discussion, which should serve as a guideline running through the whole day of the course (Figure 3). Finally, the individual methodological aspects were developed independently (30-40 minutes) by small groups (2-4 participants). For this, literature (Fitz, 1965; Adams, 1976; Karmen, Wroblewski, Ladue, 1955; Huang et al., 2006; Marienfeld Laboratory Glasware, 2017; Zander, Lang, Wolf, 1984; Keller, 1965; Pundir et Chawla, 2014; Basset et al., 1978; Wikipedia 'Ioelectric Focussing', 2017; Butler, 2012; QIAGEN DNeasy Blood&Tissue Handbook, 2017; Bray et Garnham, 1982; Wikipedia 'Malaria', 2017; Fearon, 2005; Gauthier et Turner, 1989; Narayanan et Appleton, 1980; Goldring, 2012) and orientation questions were handed out for all the experimental approaches to be performed by the students. The lecturer held individual discussions when there were questions and when required sketched out the expectations. The practical course notes provided the appropriate citations and space for notes.

Figure 3: Advance Organizer correlating symptoms, differential diagnoses and verification procedures.

This procedure was to train the expertise of the participants in selectively gathering relevant information from specialist articles, in processing it, and describing it clearly and coherently to a target group with defined background knowledge. The duration of the presentation was maximum 5 minutes for each piece of group work. The listeners should be able to understand the underlying principles, however not at the same depth of understanding as the small group that prepared the material. Either the board or a tablet PC connected by WLAN to a projector was available.
for visualization. In the end, all the participants should be able to perform all the tests using the practical course notes, and to derive the underlying mechanisms.

The laboratory diagnostics themselves were repeated in small groups. For this, eight laboratory benches were available, each equipped for a specific test (Figure 2). The amount of work required for the tests varied between 10 and 90 minutes. The students could begin with the test of their choice, depending on the area of interest, at the same time it did not need to be explicitly the test they theoretically worked on in their small group. In the available 2.5 hours of laboratory time, each small group ought to have performed several tests so that at the end of the practical day, all the tests were covered. Creatinine determination in the urine, blood glucose determination and HIV ELISA yielded normal findings. An erythrocyte count and an increase in lactate dehydrogenase in the blood indicated hemolytic anemia. Sickle-cell hemoglobin was demonstrated using agarose gel electrophoresis. Elevated protein concentrations in the urine indicated nephropathy as a result of recurring circulatory disturbances of the kidneys. To support sickle-cell anemia at a genetic level, DNA was isolated from whole blood, which was then used as the basis for amplification and sequencing (not performed here). The sequencing data were analysed more closely during the second day of the practical course. In the blood smear (here performed as an example of a Giemsa stained permanent preparation), intracellular Plasmodia were detected.

In the final seminar, all the small groups presented their primary data and explained their conclusions. The lecturer supplemented the advance organizer created at the start (Figure 3) using the test results. Positive and negative controls were included in all the experiments so that the reliability of the data could be discussed.

2.4.2 Practical day “Literature search and bioinformatic analyses” (Day 2)

Based on the diagnosis in Day 1 of sickle-cell anemia and malaria infection, the students learned to search further literature under supervision and to perform bioinformatic analyses to the molecular aspects. The second practical day (Figure 4) took place in the computer pool. During a period of free work, the students processed the tasks in the order given in the notes. The lecturer monitored the learning progress at random and supported the students with explanations, help and discussions. Each small group was assigned a task, which was to be (if necessary with additional questions) explored in depth and presented to the other small groups in the final seminar.
Figure 4: Organization of Day 2: “Literature search and bioinformatic analyses”

The task covered handling the internet portal of the National Center for Biotechnology Information and the
literature database PubMed. The students searched and skimmed over a case study about sickle-cell anemia (S/C) (O’Keeffe, Rhodes, Woodworth, 2009). They gathered information about the position of the β-hemoglobin gene (HBB) on chromosome 11 (NCBI MapViewer, 2017), its organization (introns/exons) and the known clinically-relevant mutations (NCBI Gene HBB, 2017). To be able to understand how the patient DNA isolated on Day 1 could be used for sequence clarification of the HBB gene, they repeated the working principle of the chain termination method according to Sanger (Pagel, 2017). The lecturer provided the HbS sequence digitally.

The students considered how a simple procedure for sequence comparison could be technically implemented. Furthermore, they calculated a multiple sequence comparison (diverse glucokinases, presented with CLUSTALΩ as an example, and identified the conserved regions and the relationship of the underlying organisms. They used the tool BLAST2seq to identify coding areas in the available sequences (comparison HBB wtDNA with HBB wtRNA) and to localize the disease-causing point mutation (comparison HBB wtDNA with “patient” HBS DNA). They confirmed the results through comparison with the sequencing chromatogram and examined the effects on the primary structure of the gene product (Expasy Translate).

Further tests covered the tertiary structures of the wildtype (HbA1, Protein Data Bank: 1hab) and sickle-cell hemoglobin (HbS, Protein Data Bank: 2hbs) using BALLview (BALL Project, 2017). At the same time, the students recognized how the input of additional hydrophobic areas into the protein surfaces can lead to the formation of fibrils and therefore to the sickle-cell phenotype. Additionally, they performed analyses in transmembrane domain prediction (TMHMM, 2017) and signal sequence prediction (SignalP, 2017). The specialist groups had the task of presenting the methods, which enabled in silico prediction or experimental structure determination.

The students researched why sickle-cell anemia presented a selective advantage in areas where malaria is endemic. They found out about how reestablishment of fetal hemoglobin (HbF) in adults could lead to milder symptoms of the disease (Xu et al, 2011).

In the final seminar, the specialist groups presented their results. For this, the image from every work computer could be presented using a projector. The lecturer moderated the presentation in such a way that a coherent and logical complete speech was formed from the individual presentation rounds. This process allowed the students to obtain basic information about those aspects that they themselves did not handle (e.g. through in depth work with individual questions corresponding to the personal area of interest and due to lack of time). The goal was definitely not to master all the methods in detail. Rather, Day 2 was supposed to create a founded overview of the multitude of available methods and as a result to allow the students access to bioinformatics as required.

2.4.3 Practical day “Development of a gene therapy” (Day 3)

As an introduction to Day 3 (Figure 5), the group work once again picked up on the knockout of the fetal hemoglobin repressor BCL11A (Xu et al., 2011). Building on this, in the following seminar aspects were developed that are required for understanding the development of a gene therapy. For this, the students received in small groups printouts of original publications. They covered in detail the functional principle of RNAi (Dykxhoorn et Lieberman, 2006; Eggert et Fischer, 2003), the uptake of DNA into eukaryotic cells using viral vectors (Tomar, Matta, Chaudhary, 2003), the benefits and risks of gene therapies (Wagenmann, 2017; Böhme, Dörner, Ehrhardt, 2017), erythropoiesis (Smith, 2003) (and with it topism / the route of application of a potential drug) and the functionality of the pAdEasy™ system (Khatun, 2012). The duration of the presentation was a maximum of 5 minutes for each piece of group work with an additional 2 minutes for discussion. In a subsequent group phase, the learned aspects were integrated by the lecturer and the concept presented for the development of a gene therapy: Adenoviruses could be used to infect the erythroblasts of the bone marrow, to deliver a plasmid coding for short hairpin RNA (shRNA). This shRNA would include a sequence fragment of the fetal hemoglobin repressor BCL11A and would be processed into small interfering RNA (siRNA) in the erythroblasts. In doing so, the expression of
BCL11A would be downregulated by RNAi, which would lead to the production of fetal hemoglobin, thus counteracting the tendency of HbS to form fibrils. Care must be taken that the virus used is itself not able to replicate. The infection of erythroblasts could take place for example *in vitro* following bone marrow biopsy and the transgenic cells could be reimplanted after chemotherapy.
Re-entry and discussion of the objectives
The last specialist group of Day 2 restates their results. The lecturer defines the objectives of the development of a gene therapy as knockdown of fetal hemoglobin repressor BCL11A.

Group work
- RNA interference (Dykshoorn, 2006; Eggert, 2003)
- adeno-/retroviruses as a means to deliver DNA, (dis-) advantages (persistence / oncogenes) (Tomar, 2003)
- benefits and risks of gene therapies I (Wagenmann, 2017)
- benefits and risks of gene therapies II (Böhme, 2017)
- erythropoiesis (Smith, 2003), route of application (biopsy of bone marrow, in vitro-modification, chemotherapy necessary?, reimplantation)
- use of the pAdEasy™-system (Khatun, 2012)

Presentation of group work
(aim: comprehension of the fundamental principles, max. 5 minutes/group plus 2 minutes moderated discussion)
Available media: blackboard, interactive whiteboard (tablet PC with WLAN connection to beamer)

Presentation of the cloning concept by the lecturer
• cloning (to be performed during the practical)
  o restriction of pSilencer™ with BamH I / HindIII
  o ligation of pSilencer-BCL11A* (codes for shRNA and contains BbsI restriction site)
  o transformation in E.coli, selection with ampicillin
  o picking clones, inoculation of 3ml cultures
  o isolation of DNA (miniprep)
  o restriction analysis with BbsI
  o agarose gel electrophoresis
• homologous recombination in BJ5183 (in theory)
  o co-transformation of pSilencer™-BCL11A* and pAdEasy™-1 in BJ5183
  o selection (kanamycin) and picking the smallest clones, 3ml culture
  o isolation of DNA (miniprep)
  o linearization with PstI
  o restriction analysis / gel electrophoresis
• production of viruses in AD-293 (in theory)
  o clean-up (removal of PstI)
  o transfection of AD-293
  o harvesting viruses
• outlook
  o infection of bone marrow cells
  o control of BCL11A expression level by qPCR/Northern blot
  o compatibility study in animal model
  o trial of effectiveness in animal model
  o infection of bone marrow cells (patient), chemotherapy, reimplantation

Performance of assays in small groups (approx. 3 persons).
Eight work benches with a full set of pipettes/chemicals are available for each distinct cloning step. The small groups start at staggered times (every 10 minutes) and perform the tasks in the given order. Alternating small groups perform the cloning steps 1-4 (Block 1) or 4-8 (Block 2).

The lecturer gives the first small groups of each block an instruction, which should be passed on by the students in succession to the subsequent small groups. The lecturer checks at regular intervals whether all the groups are sufficiently informed.

Block 1
- restriction of pSilencer™ with BamH I / HindIII
- preparation of LB-agar-ampicillin-plates
- ligation of pSilencer-BCL11A* (codes for shRNA und contains BbsI restriction site)
- transformation in competent E.Coli, selection with ampicillin

Block 2
- picking clones, inoculation of 3ml cultures
- isolation of DNA (miniprep)
- restriction analysis with BbsI
- agarose gel electrophoresis

Logging directly into designated fields of the course notes.
(free space for observations or comments)

Preparation of Day 4 (piece of homework: each small group should prepare one paragraph of a manuscript. The following keywords are given in the practical notes):

- Abstract. Short summary of the entire project: case study, sickle cell anemia, co-infection with malaria, RNAi of BCL11A, adeno viral vectors, possible therapeutic agent to be injected into the bone marrow
- Introduction 1. Sickle cell anemia / malaria: prevalence, distribution, expectation of life
- Introduction 2. Existing publication concerning BCL11A, RNA interference, pAdEasy™-system
- Methods 1. Erythrocyte count, detection of plasmmodia (HE-stain), Hb-electrophoresis
- Methods 2. LDH-determination in blood, protein determination in urine
- Methods 3. Cloning of pSilencer™- BCL11A*
- Results 1. Anamnesis
- Results 2. Anemia (RBC count), Hb-electrophoresis, LDH (diagnosis)
- Results 3. Protein concentration in urine, plasmmodia (diagnosis)
- Results 4. Cloning of pSilencer™ (results of restriction analysis)
- Discussion 1. Patient diagnosis (short), description of pAdEasy™-system, cloning (short), outlook: recombination with pAdEasy™-1
- Discussion 2. Outlook: production of viruses, infection of erythroblasts, measurement of expression level (qPCR)
Figure 5: Organization of Day 3: “Development of a gene therapy”

In practice, on Day 3 an oligonucleotide (BCL11A*) should be used, which contains a 19 nucleotide-long fragment from BCL11A in its sequence, followed by a hairpin structure (recognition site of BbsI), and the reverse complementary sequence fragment of BCL11A*. 5’-terminal found a recognition site for BamHI, 3’-terminal for HindIII. The oligonucleotide should be cloned in the vector pSilencer™ 2.1-U6-Hygro and the correct integration confirmed using a restriction control (BbsI).

The follow-on procedure was discussed in the seminar, but was not part of the experiments: pSilencer™-BCL11A* would be co-transformed into BJ5183 using pAdEasy™-1. Through homologous recombination, pAdEasy™1-1-pSilencer™-BCL11A* would be formed and isolated. This bacmid could be transfected in AD-293 cells. The AD-293 cells are human embryonic kidney cells (HEK), which express the protein essential for viral reproduction in trans. Thus, they enable the multiplication of adenoviruses (which lack this protein) using vectors such as pAdEasy™-1. Viruses would be formed that cannot replicate and therefore did not represent a risk of infection for the patients.

The experimental procedure on Day 3 took place in small groups, which each ran through the first or second block of the cloning in the given order (Figure 5). The small groups began at intervals of 10 minutes. The lecturer gave the first small group an instruction, which was passed on by the students in succession to the subsequent small groups. The lecturer checked at regular intervals whether all the groups were sufficiently informed.

For the sub-steps of cloning, which usually lead to long waiting periods (like for example the cultivation of E.coli over night), analogously produced samples were made available. These were either prepared by the lecturer himself, of taken over from the last practical group.

After the end of the event, in addition to being able to perform the test adequately, the students should also be able to carry out independently the calculations necessary for its preparation. This covers, for example, volume calculation for restriction / ligation preparations, the dilution of concentrated stock solutions, or the calculation of the quantities of materials to be weighed out where percentage amounts are given. Questions / indicators and space for notes (or gaps in the text) were available in the practical notes.

2.4.4 Practical day “Publication and reflection” (Day 4)

Before the start of the last practical day, the students created individual sections of a manuscript in small groups (Figure 5). The aim of the topic was made known during the previous practical day. The submitted parts of the publication were combined by the lecturer to form a finished manuscript and the title, authors, figures (e.g. gel electrophoresis, calibration curves and restriction analysis) and references added. The volume of the combined document should not exceed five A4 pages. The students should learn to formulate their information as briefly and precisely as possible (e.g. by giving the concentrations of the used reagents or literature citations instead of an extensive description of the standard procedures). In the framework of a seminar, the small groups presented their part of the manuscript, during which measurement errors or measurement inaccuracy, completeness and reproducibility were critically scrutinized. It was negated in a moderated discussion how publishable the manuscript was, and the prospects were compiled concerning which further steps were still needed for successful development and testing of the gene therapy (at least as far as necessary for successful publication). BCL11A knockdown using shRNA was still a component of real research when the practical experiment was designed and during the pilot trials in the 2016 summer semester. Anyhow, this therapeutic approach was published in the meantime. The students of later practical courses were informed of this and received a copy of the respective publication (Brendel et al., 2016).
3. Results

The practical course was evaluated in its original format (from summer semester 2011 to summer semester 2016) with an average overall grade of 2.82. The newly designed practical (from summer semester 2016 to winter semester 2017/18) with an average overall grade of 1.62. These data were routinely collected by the medical faculty using the evaluation software tuevalon (Ostrakon Software GmbH) (Figure 1).

In the summer semester of 2016, both practical courses took place simultaneously, whereby the six practical groups were randomly assigned (old course: 57 participants, new course: 79 participants). Separate questionnaires were compiled for comparative evaluation, which were filled out anonymously by the students at the end of the event. The evaluation covered the educational concept (Figure 6a), degree of difficulty (6b), learning achievement (6c) and overall evaluation (6d), organizational structure (6e), focal points (6f), quality of the course materials (6g) and supervision (6h), comprehensibility (6i) and integration in the greater context (6j), as well as relevance for the medical profession (6k, 6l), acquisition (6m) and transfer of knowledge (6n).

The students quoted an almost unchanged degree of difficulty (Figure 6b: I found the level of difficulty to be…; old: 3.2 “exactly right”; new: 3.1 “exactly right”) for obtaining greater learning achievement (Figure 6c: My personal learning achievement is…; old: 3.2 “satisfactory”; new: 1.8 “good”). They were better able to recognize the context of the experimental content (Figure 6j; I could integrate the learned material into the greater context; old: 3.3 “undecided”; new: 1.7 “agree somewhat”). Furthermore, the importance of the material was more apparent (Figure 6k: I find the learned content to be…; old: 3.2 “undecided”; new: 1.9 “important”) for working in the medical profession and the NKLM role of scholars (Figure 6m: The event has helped me to expand my knowledge independently in the future; old: 3.0 “undecided”; new: 1.8 “agree somewhat”) and communicators (Figure 6n: The event has helped me to convey my knowledge intelligibly to other people; old: 3.5 “disagree somewhat”; new: 2.1 “agree somewhat”) were covered better. The students evaluated the original practical course with the overall grade 2.9, the newly designed practical course with the overall grade 1.4 (Figure 6d: I give the event the overall grade…).
Figure 6: Comparative evaluation of both practical courses in summer semester of 2016. n(old)=57; n(new)=79.

4. Discussion

The author is convinced that the key task of a biochemistry practical course remains to impart specialist information, as stated in the NKLM (NKLM, 2015) under “Principles of normal structure and function” (NKLM, Chapter 12) and “Principles of pathogenesis and pathomechanisms” (NKLM, Chapter 13), and are more precisely defined in the content outline of the Institute’s medical and pharmaceutical examination questions (IMPP). However, it was shown here that the preclinical practical is also suitable for training further roles of the medical practitioner early on in the curriculum, without reducing the specialist content (Figure 7). In the practical course presented here, the specialist content was treated using a concrete case example so that the students were already compelled in the preclinical section to plan a suitable diagnostic for their patient, to identify the relevant aspects of the given symptoms, and to take into consideration the ethnic background (NKLM, Medical expert, chapter 5). At the same time, the ethical aspects and risks to which the patient could potentially be exposed were discussed (NKLM, Professional doer, chapter 11). By working through individual topic areas using scientific publications, as well as through independent literature reviews, the students should be prepared for independent continued training throughout life (NKLM, Scholar, chapter 6). Group work aimed to strengthen the organizational ability and personal
responsibility, and to present the learned material in an appropriate manner (NKLM, Member of a team, chapter 8). With various experiments, which took place in an overall context, and the corresponding recording and collation of a manuscript, the basis of the scientific work was conveyed to the students (NKLM, Medical-scientific skills, chapter 14a). Furthermore, technical understanding, ability to observe and bioinformatic basic knowledge were to be trained.

Figure 7: Overview over the topics of practical days 1-4 and coverage of the physician roles according to the German National Competence-based Learning Objectives Catalogue for Undergraduate Medical Education (NKLM)

In a comparative evaluation, the students graded their learning achievement as significantly better than with traditional teaching formats with lecturer-based seminars as preparation for the practical and classical biochemical tests without a greater context. At the same time, the relevance of the taught content was more apparent to the students.

The presented concept (and the comparison practical course) was carried out with a supervision ratio of 25:1. For this, the around 140-150 students in one semester were split into 6 groups. In the newly designed practical course, each group was supervised by one lecturer in all the sequential experimental days, and the participants received continuous individual support. On the organizational level, the individual groups were staggered at the start of the practical course so that at least two lecturers were required for supervision (each for 12 days) of all the semester cohort. In the comparison practical, four lecturers always supervised the same experimental day but with different practical groups (in each case six days). Both practical courses took place as a 3-week block event. In this way, the personnel requirements appear manageable and are not necessarily limiting for the adaptation of existing teaching methods to new curricular developments. It is expected that the running costs for consumables and chemicals remain constant after the redesign of the practical course.

5. Conclusions

In summary, it was shown that despite existing financial, personnel and time restrictions, an adjustment of the biochemistry practical course to the requirements of the NKLM roles of medical practitioners was possible, and that this development was supported by the students. Commitment increased through the recognizable clinical and scientific reference. According to our observations, training in the additional roles of medical practitioners does not mean a reduction in specialist teaching content, but rather an increase in motivation. We are convinced that the concept will lead in the long-term to an improvement in the learning success. During the next years, nearly all
subjects of the preclinical section will be challenged to find ways of combining NLKM requirements and hard facts. This study can serve as a model to renew practical concepts, as the here presented didactics can be easily adopted to other fields like physiology, anatomy, physics or biology.

Take Home Messages

- Medical students often have difficulties to identify the relevance of basic biochemistry to their profession.
- Putting practical courses into a context with recognizable clinical and scientific relevance increases commitment.
- Working in specialist groups strengthens the students organizational ability and personal responsibility.
- Training in the additional roles of medical practitioners (as set out in the German National Competence-based Learning Objectives Catalogue for Undergraduate Medical Education) does not mean a reduction in specialist teaching content, but rather increases the students motivation.

Notes On Contributors

Stefan Mogk studied biochemistry at the University of Tübingen, Germany. In his doctoral thesis he investigated the brain infection of African Sleeping Sickness. Since 2013 he has coordinated biochemical education of medical students at the University of Tübingen. Since winter 2016/17 he has lectured basic biochemistry for medicals in the 2nd semester. His readings have been awarded with the Tuevalon Teaching Price 2018 of the medical faculty.

Acknowledgements

The author would like to thank Prof. Dr. Thorsten Stafforst and Alfred Hanswillemenke for general assistance, as well as Jan Griewatz and Dr. Axel Strauß for writing assistance.

Bibliography/References

Adams, P. (1976) 'The Tetramerisation of LDH subunits in human serum.', Int. J. Biochem., 7, pp. 515-517. https://doi.org/10.1016/0020-711X(76)90054-9

BALL Project. Eberhard Karls Universität Tübingen. Available at: https://ball-project.org/ (Accessed: 14.08.2017).

Basset, P., Beuzard, Y., Garel, M. C. and Rosa, J. (1978) 'Isoelectric focusing of human hemoglobin: its application to screening, to the characterization of 70 variants, and to the study of modified fractions of normal hemoglobins', Blood, 51(5), pp. 971-82.

Böhme, P., Dörner, J. and Ehrhardt, A. (2015) Gentherapie - längst keine Utopie mehr. Available at: https://www.pharmazeutische-zeitung.de/index.php?id=60690 (Accessed: 14.08.2017).

Bray, R. S. and Garnham, P. C. (1982) 'The life-cycle of primate malaria parasites', Br Med Bull, 38(2), pp. 117-22. https://doi.org/10.1093/oxfordjournals.bmb.a071746

Brendel, C., Guda, S., Renella, R., Bauer, D. E., et al. (2016) 'Lineage-specific BCL11A knockdown circumvents toxicities and reverses sickle phenotype', J Clin Invest, 126(10), pp. 3868-3878. https://doi.org/10.1172/JCI87885
Butler, J. (2012) 'DNA Extraction Methods (Chapter 2). ', Advanced Topics in Forensic DNA Typing: Methodology.

Dykxhoorn, D. M. and Lieberman, J. (2006) 'Knocking down disease with siRNAs', Cell, 126(2), pp. 231-5. https://doi.org/10.1016/j.cell.2006.07.007

Eggert, C. and Fischer, U. (2003) 'RNA-Interferenz: Ein neues Werkzeug zur Analyse der Genfunktion', BIOSPEKTRUM Heidelberg, 9, pp. 372-374.

Fearon, M. (2005) 'The laboratory diagnosis of HIV infections', Can J Infect Dis Med Microbiol 16(1), pp. 26-30. https://doi.org/10.1155/2005/515063

Fritz, P. J. (1965) 'Rabbit muscle lactate dehydrogenase 5; a regulatory enzyme', Science, 150(3694), pp. 364-6. https://doi.org/10.1126/science.150.3694.364

Gauthier, D. K. and Turner, J. G. (1989) 'Anti-HIV antibody testing: procedures and precautions', Am J Infect Control, 17(4), pp. 213-25. https://doi.org/10.1016/0196-6553(89)90131-4

Gene HBB (NCBI Gene hemoglobin subunit beta). Available at: https://www.ncbi.nlm.nih.gov/gene/3043 (Accessed: 14.08.2017).

Goldring, J. P. (2012) 'Protein quantification methods to determine protein concentration prior to electrophoresis', Methods Mol Biol, 869, pp. 29-35. https://doi.org/10.1007/978-1-61779-821-4_3

Huang X, Yang-Kyu C, Hyung-Soon I, Oktay Y., et al. (2006) 'Aspartate Aminotransferase (AST/GOT) and Alanine Aminotransferase (ALT/GPT) Detection Techniques', Sensors 6, pp. 756-782. https://doi.org/10.3390/s6070756

Karmen, A., Wroblewski, F. and Ladue, J. S. (1955) 'Transaminase activity in human blood', J Clin Invest, 34(1), pp. 126-31. https://doi.org/10.1172/JCI103055

Keller, D. M. (1965) 'An Enzymatic Method for Glucose Quantitation in Normal Urine', Clin Chem, 11, pp. 471-7.

Khatun, I. (2012) 'Recombinant Adenoviral Vector Systems ', Material and Methods, 2, p. 126. https://doi.org/10.13070/mm.en.2.126

MapViewer. NCBI MapViewer chromosome 11. Hemoglobin subunit beta. No longer available (has been replaced by Genome Data Viewer).

Marienfeld Laboratory Glassware. Zählkammer. Available at: https://seidel-medipool.medizin-logistik.com/shop_images/Download/2010-Marienfeld-info-Zaehlkammern.pdf (Accessed: 14.08.2017).

Narayanan, S. and Appleton, H. D. (1980) 'Creatinine: a review', Clin Chem, 26(8), pp. 1119-26.

NKLM (2015) Nationaler Kompetenz-basierter Lernzielkatalog Medizin. Available at: www.nklm.de (Accessed: 14.08.2017).

O’Keeffe, E. K., Rhodes, M. M. and Woodworth, A. (2009) 'A patient with a previous diagnosis of hemoglobin S/C disease with an unusually severe disease course', Clin Chem, 55(6), pp. 1228-31; discussion 1231-3. https://doi.org/10.1373/clinchem.2008.112326

Pagel, P. Genomsequenzierung für Anfänger. Available at: http://docplayer.org/3967213-Genomsequenzierung-fuer-anfaenger.html (Accessed: 14.08.2017).

Pundir, C. S. and Chawla, S. (2014) 'Determination of glycated hemoglobin with special emphasis on biosensing methods', Anal Biochem, 444, pp. 47-56. https://doi.org/10.1016/j.ab.2013.09.023
QIAGEN DNeasy Blood & Tissue Handbook. Available at: https://www.qiagen.com/de/resources/resourcedetail?id=6b09dfb8-6319-464d-996c-79e8c7045a50&lang=en (Accessed: 14.08.2017).

SignalP Technical University of Denmark. Center for biological sequence analysis. Prediction of signal sequences. Available at: http://www.cbs.dtu.dk/services/SignalP/ (Accessed: 14.08.2017).

Smith, C. (2003) 'Hematopoietic stem cells and hematopoiesis', Cancer Control, 10(1), pp. 9-16. http://dx.doi.org/10.1177/107327480301000103

Thiel, M. (2013) Modellstudiengänge für Medizin. Available at: https://www.thieme.de/viamedici/vor-dem-studium-infos-zum-medizinstudium-1493/a/modellstudiengaenge-medizin-3737.htm (Accessed: 14.8.2017).

TMHMM Technical University of Denmark. Center for biological sequence analysis. Prediction of transmembrane helices in proteins. Available at: http://www.cbs.dtu.dk/services/TMHMM/ (Accessed: 14.08.2017).

Tomar, R. S., Matta, H. and Chaudhary, P. M. (2003) 'Use of adeno-associated viral vector for delivery of small interfering RNA', Oncogene, 22(36), pp. 5712-5. http://dx.doi.org/10.1038/sj.onc.1206733

Wagenmann, U. Gentherapie erfolgreich, Patient tot? Gen-ethisches Netzwerk e.V. Berlin. Available at: http://gen-ethisches-netzwerk.de/gentherapie-erfolgreich-patient-tot (Accessed: 14.08.2017).

Wikipedia Isoelectric focusing. Available at: https://en.wikipedia.org/wiki/Isoelectric_focusing (Accessed: 14.08.2017).

Wikipedia Malaria. Available at: https://de.wikipedia.org/wiki/Malaria (Accessed: 14.08.2017).

Wissenschaftsrat (2014) Empfehlungen zur Weiterentwicklung des Medizinstudiums in Deutschland auf Grundlage einer Bestandsaufnahme der humanmedizinischen Modellstudiengänge. Available at: https://www.wissenschaftsrat.de/download/archiv/4017-14.pdf (Accessed: 14.08.2017).

Xu, J., Peng, C., Sankaran, V. G., Shao, Z., et al. (2011) 'Correction of sickle cell disease in adult mice by interference with fetal hemoglobin silencing', Science, 334(6058), pp. 993-6. https://dx.doi.org/10.1126%2Fscience.1211053

Zander, R., Lang, W. and Wolf, H. U. (1984) 'Alkaline haematin D-575, a new tool for the determination of haemoglobin as an alternative to the cyanhaemiglobin method. I. Description of the method', Clin Chim Acta, 136(1), pp. 83-93. https://doi.org/10.1016/0009-8981(84)90250-X

Appendices

None.

Declarations

The author has declared that there are no conflicts of interest.

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Ethics Statement

The author hereby declares that all data were collected in strict accordance with local law and the Declaration of Helsinki. Student evaluation data was acquired on a voluntary and anonymous basis for quality assurance as prescribed in the University Law of Baden-Württemberg (Germany). This was also confirmed by the ethics committee of the University Hospital Tübingen.

External Funding

This project was financed by the medical faculty of the Eberhard-Karls-University of Tübingen (PROFILplus).