

## Kvaliteta slika elektroforeze i izoelektričnog fokusiranja komprimiranih primjenom JPEG i JPEG2000

### Quality of electrophoresis and isoelectric focusing images compressed using JPEG and JPEG2000

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#### Sažetak

**Cilj:** Za potrebe telepatologije mora se pri prijenosu zadržati visoka kvaliteta svih elemenata slike. Cilj je ovog rada bio komprimirati slike uzoraka elektroforeze i izoelektričnog fokusiranja algoritmima JPEG (engl. *Joint Photographic Expert Group*) i JPEG2000 te procijeniti kvalitetu komprimiranih slika prije i poslije elektronskog prijenosa.

**Metode:** Skenogrami uzoraka elektroforeze serumskih proteina bolesnika s albuminom Zagreb i izoelektričnog fokusiranja albumina bolesnika s albuminom Krapina te fotografirani skenogram izoelektričnog fokusiranja odabrani su za istraživanje. Svaka je slika komprimirana u osam stupnjeva (od 3,0 bpp (engl. *bit per pixel*, bit po pikselu) do 0,1 bpp) primjenom kompresijskih postupnika JPEG i JPEG2000. Sve slike ( $N = 51$ ), komprimirane i nekomprimirane, poslane e-poštom, procjenjivalo je osam medicinskih biokemičara: šest iz Hrvatske, jedan iz Italije i jedan iz Danske. Kvaliteta slika vrednovana je i objektivnim mjerilima kvalitete slika, tj. PSNR (engl. *Peak-Signal-to-Noise-Ratio*, omjer vršnog signala i šuma), SNR (engl. *Signal-to-Noise-Ratio*, omjer signala i šuma), OQF (engl. *Optimized Quality Factor*, optimirani činitelj kvalitete) i MSE (engl. *Mean Squared Error*, glavna kvadratna pogreška).

**Rezultati:** Subjektivnim ocjenjivanjem sve su slike komprimirane algoritmom JPEG2000 ocijenjene kao *izvrsne*. Protivno tome, kompresijom JPEG pri kompresijama od 0,1 bpp slike su ocijenjene kao *potpuno neupotrebljive*, kompresijom od 0,2 bpp ocijenjene su kao *dobre ili umjereno nejasne*, a slike komprimirane od 0,3 do 3,0 bpp ocijenjene su kao *izvrsne*. Vrijednosti PSNR i SNR kod kompresije JPEG od 0,3 bpp bile su sukladne vrijednostima PSNR i SNR kod kompresije JPEG2000 od 0,1 bpp.

#### Abstract

**Aim:** High quality of all image elements must be maintained for the purpose of telepathology. The aim of this study was to compress the images of electrophoresis and isoelectric focusing samples using JPEG (*Joint Photographic Expert Group*) and JPEG2000 algorithms, and to assess the quality of the compressed images before and after electronic transmission.

**Methods:** Scanograms of serum protein electrophoresis samples of a patient with Zagreb albumin and albumin samples for isoelectric focusing of a patient with Krapina albumin were selected for the study together with a photographed scanogram of isoelectric focusing. Each image was compressed at eight compression rates (from 3.00 bpp (*bit per pixel*) to 0.1 bpp) using JPEG and JPEG2000 compression algorithms. All images ( $N = 51$ ), both compressed and uncompressed, were transmitted by email for assessment to eight medical biochemists: six from Croatia, one from Italy and one from Denmark. Image quality was also assessed by objective measures, i.e. compared to the quality of PSNR (*Peak-Signal-to-Noise-Ratio*), SNR (*Signal-to-Noise-Ratio*), OQF (*Optimized Quality Factor*) and MSE (*Mean Squared Error*) images.

**Results:** All images compressed using the JPEG2000 algorithm were subjectively rated as *excellent*. Contrarily, images compressed using JPEG at 0.1 bpp were rated as *completely useless*, those at 0.2 bpp as *moderately blurred*, and those at 0.3–3.00 as *excellent*. At JPEG compression at 0.3 bpp, PSNR and SNR values corresponded to PSNR and SNR values obtained by JPEG2000 compression at 0.1 bpp.

**Zaključak:** Nakon elektronskog prijenosa subjektivna ocjena kvalitete slika komprimiranih algoritmom JPEG2000 od 0,1 do 3,0 bpp, te slika komprimiranih algoritmom JPEG od 0,3 do 3,0 bpp bila je jednaka kvaliteti izvornih slika.

**Ključne riječi:** JPEG, JPEG2000, kompresija slika, analiza slika, telemedicina, elektroforeza, izoelektrično fokusiranje

**Conclusion:** According to subjective rating, the quality of images compressed by using JPEG2000 algorithm at 0.1-3.0 bpp and JPEG algorithm at 0.3-3.0 bpp was equal after electronic transmission to the quality of original images.

**Key words:** JPEG, JPEG2000, image compression, image analysis, telemedicine, electrophoresis, isoelectric focusing

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## Uvod

Telemedicina, koja je udružila dvije specijalnosti, tj. medicinu i telekomunikacije, omogućila je brzi elektronski prijenos laboratorijskih slika (engl. *store-and-forward telemedicine*) u svrhu postavljanja primarne dijagnoze, dobivanje drugog mišljenja specijalista, u svrhu profesionalne izobrazbe i usavršavanja, dugotrajne pohrane podataka u digitalnom obliku, te za potrebe video-konferencija (tj. interaktivna primjena u realnom vremenu) (1). Slike se u laboratorijskoj medicini obično prenose kao slike snimljene digitalnom kamerom, a u zadnje vrijeme kao slike dobivene mikroskopima s digitalnom opremom. Digitalizirane slike moraju imati veliku razlučivost (2,3). U složenom procesu digitalizacije, kompresije i elektronskog prijenosa slike, kvaliteta izvorne slike mora biti u cijelosti očuvana. Kako se kompresijom ne bi izgubila visoka razlučivost izvorne slike, moraju se primijeniti kvalitetni algoritmi za kompresiju slike uz veliki kapacitet telefonske mreže (4). JPEG (engl. *Joint Photographic Expert Group*) je zajedničko ime za radnu skupinu koja je razvila postupak kompresije mirnih slika. Razvoj, vrednovanje i standardizacija trajali su od 1982. do 1987. godine, a 1992. godine JPEG skupina je dovršila opis postupka za kompresiju fotografija sa i bez gubitaka (engl. *lossy and lossless*) koji je prihvaćen kao međunarodni standard (5). JPEG2000 je novi međunarodni standard temeljen na tehnologiji *wavelet* (6). Algoritam JPEG 2000 može u cijelosti ili djelomično kodirati datoteke bez gubitaka, pa je pogodan za obradu medicinskih slika (6). Bez obzira koji se kompresijski algoritam primjenjuje, važno je ocijeniti do kojeg se stupnja slika može komprimirati, a da ne izgubi na kvaliteti koja je nužna za njezino vrednovanje (7). Kvaliteta slike ocjenjuje se subjektivnim i objektivnim mjerilima.

Cilj je ovog rada bio komprimirati slike uzoraka elektroforeze i izoelektričnog fokusiranja algoritmima JPEG i JPEG2000 te procijeniti kvalitetu komprimiranih slika prije i poslije elektronskog prijenosa. Slike elektroforeze i izoelektričnog fokusiranja bile su upotrijebljene u multicentričnoj evaluaciji bolesnika s bisalbuminemijom.

## Introduction

Telemedicine, which has integrated two specialties, i.e. medicine and telecommunications, has enabled rapid electronic transmission of laboratory images (*store-and-forward telemedicine*) for various purposes: making initial diagnosis, obtaining a specialist's second opinion, professional training and education, long-term data storage in digital format, and videoconferences (interactive application in real time) (1). Images are in laboratory medicine usually transferred as images taken by digital camera, and lately also as images obtained on microscopes with digital accessories. Digitized images must have high resolution (2,3). In a complex process of digitalization, compression and electronic image transmission, the quality of the original image must be entirely retained. In order not to lose high resolution of an original image by compression, quality algorithms must be applied for image compression along with a high capacity telephone network (4). JPEG (Joint Photographic Expert Group) is a common name for a working group that has developed a procedure to compress still images. Development, evaluation and standardization were carried out from 1982 to 1987, and in 1992 the JPEG Group finalized the specification of procedures for lossy and lossless compression of tone still images that was accepted as a national standard (5). JPEG 2000 is a new international standard based on the wavelet technology (6). JPEG 2000 algorithm can entirely or partially code databases without data loss, which makes it suitable for processing of medical images (6). Regardless of what compression algorithm is used, it is important to determine the highest degree of image compression without loss in quality which is necessary for image evaluation (7). Image quality is rated according to subjective and objective criteria.

The aim of this paper was to compress the images of electrophoresis and isoelectric focusing samples by JPEG and JPEG 2000 algorithms, and assess the quality of compressed images before and after their electronic transmission. Electrophoresis and isoelectric focusing images had been used in a multicentric evaluation of patients with bisalbuminemia.

## Materijali i metode

### Dobivanje digitalnih slika i kompresija

Tri su slike odabrane za ispitivanje: skenogram elektroforeze serumskih proteina na celogelu bolesnika s albuminom Zagreb (Slika 1.A1), dva uzorka izoelektričnog fokusiranja albumina Krapina na poliakrilamidu (Slika 1.B1), te fotografija uzoraka izoelektričnog fokusiranja albumina sa standardima na poliakrilamidu (Slika 1.C1). Slike su korištene u elektronskom dopisivanju pri objavljivanju rezultata istraživanja dvostrukе albuminemije (8,9). Razlučivost izvornih slika iznosila je 1199 dpi (slike 1.A i 1.B), odnosno 99 dpi (slika 1.C). Odabrane su slike skenirane (upotrebom plosnatog skenera) i potom pohranjene kao slike u računalu (veličina konvertiranih slika bila je 225–649 KB; broj elemenata 77–221 kilopiksela). Nakon pohrane, slike su konvertirane u 24-bit BMP (BMP je standardni grafički format za MS Windows; obično se primjenjuju mape s razlučivošću 24 bita po pikselu jer za polikromatske slike daje bolju kvalitetu izvorne slike) i komprimirane pomoću edukacijskog programa VCDemo, version 5.03 (10). Pri kompresiji su ispitana dva različita postupnika za kompresiju slika, tj. JPEG (5) i JPEG2000 (6). Elferogrami su komprimirani u osam različitih stupnjeva objema kompresijskim metodama, tj. 3,0 bpp (engl. *bit per pixel*, bit po pikselu), 1,5 bpp, 1,0 bpp, 0,7 bpp, 0,5 bpp, 0,3 bpp, 0,2 bpp i 0,1 bpp. Na kraju, izvorne i komprimirane slike poslane su u pravitu e-poštom različitim osobama.

### Mjerila kvalitete

Kvaliteta komprimiranih i proslijedjenih slika ispitana je subjektivnim i objektivnim načinom, uključujući (i) tri izvorne slike (Slike 1.A1, 1.B1 i 1.C1) i sve komprimirane slike ( $3 \times 17 = 51$  slika). Slike je procjenjivalo osam medicinskih biokemičara iz pet različitih ustanova: šest iz Hrvatske, jedan iz Italije (L.M.) i jedan iz Danske (U. K-H.). Svi osim L.M. i U.K-H. nisu znali kojim su algoritmom slike bile komprimirane. Svi su procjenitelji bili upoznati s elementima važnima za procjenu vrste slika i zamoljeni su da usporede komprimirane slike s izvornim slikama te ih ocijene kao: *izvrsna* (jednake kvalitetu kao i izvorna slika; nepromijenjena); *dobra ili umjerenog nejasna* (neznatno promijenjena), ili *potpuno neupotrebljiva* (potpuna dekompozicija slike; gubitak relevantnih podataka); (ii) nekomprimirane i komprimirane slike objektivno su procijenili neovisni procjenitelji M.D. i S.G. koji su znali kojim su protokolom slike bile komprimirane.

Ocjena slika učinjena je prije i poslije kompresije primjenom softvera za analizu slika koji može odrediti omjer signala i šuma (SNR, engl. *Signal-to-Noise-Ratio*), vršni omjer signala i šuma (PSNR, engl. *Peak-Signal-to-Noise-Ratio*), srednju kvadratnu pogrešku (MSE, engl. *Mean Squared Error*) i optimirani činitelj kvalitete (OQF, engl. *Optimized Quality Factor*). Među tim parametrima, MSE i PSNR su

## Materials and methods

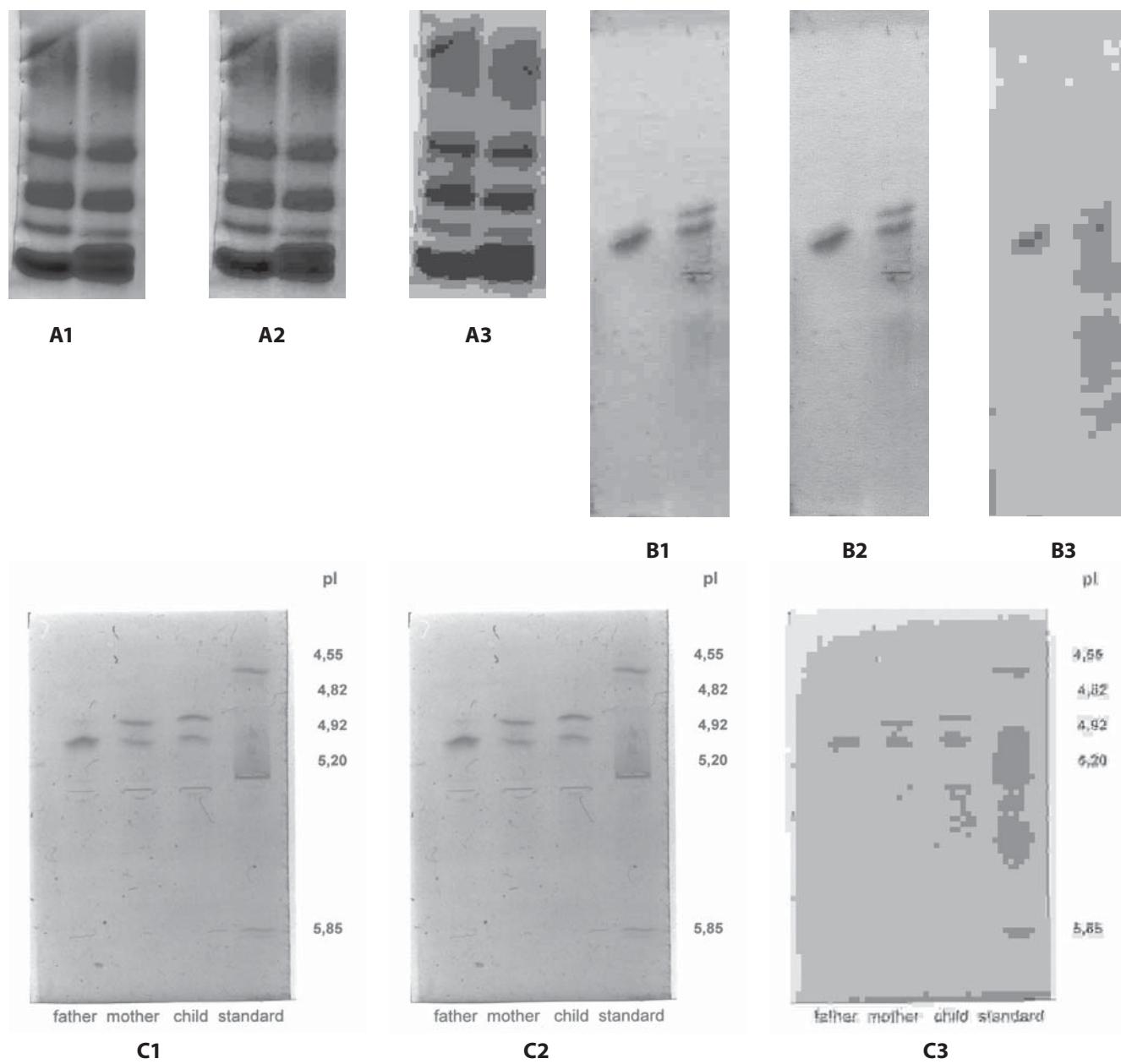
### Formation of digital images and compressions

Three images were selected for the study: a scanogram of cellogel electrophoresis of serum proteins of a patient with the Zagreb albumin (Fig. 1.A1), two samples of polyacrylamide isoelectric focusing of the Krapina albumin (Fig. 1.B1), and a photograph of polyacrylamide isoelectric focusing of albumin samples with standards (Fig. 1.C1). The images were used in email correspondence related to publication of results of a study of double albuminemia (8,9). The resolution of original images was 1199 dpi (Figures A and B) and 99 dpi (Figure C). The selected images were scanned (using a flat scanner) and subsequently stored as computer images (the size of converted images was 225–649 KB; the number of pixels was 77–221 kilopixels). After storage, the images were converted to 24-bit BMP (BMP is a standard graphic format for MS Windows; folders with 24 bits per pixel are usually applied as this ensures better quality of the original image for polychromatic images) and compressed using VCDemo educational application, version 5.03 (10). During compression, two different algorithms for image compression were tested, i.e. JPEG (5) and JPEG2000 (6). Electropherograms were compressed at eight different degrees by both compression methods, i.e. at 3.0 bpp (bits per pixel), 1.5 bpp, 1.0 bpp, 0.7 bpp, 0.5 bpp, 0.3 bpp, 0.2 bpp and 0.1 bpp. Finally, original and compressed images were emailed as attachments to different persons.

### Quality measures

The quality of compressed and emailed images was tested in a subjective and objective manner: (i) three original images (Figs 1.A1, 1.B1, 1.C1) and all compressed images ( $3 \times 17 = 51$  images) were examined. The images were rated by eight medical biochemists from five different institutions: six from Croatia, one from Italy (L.M.) and one from Denmark (U.K.-H.). Except for L.M. and U.K.-H., others were not aware of the algorithm used for image compression. All assessors were familiar with the elements important for assessing image type, and they were asked to compare the compressed images with the original ones and rate them as excellent (of equal quality to the original image; unaltered), good or moderately blurred (slightly changed), or completely useless (total image degradation; loss of relevant data); (ii) uncompressed and compressed images were objectively rated by independent assessors M.D. and S.G. who were aware of the protocols used for image compression.

The rating of images was carried out before and after compression by using software for image analysis that can determine signal-to-noise ratio (SNR), peak signal-to-noise ratio (PSNR), mean squared error (MSE) and optimized quality factor (OQF). Among the above parameters,

**SLIKA 1.**

**(A) ELEKTROFOREZA** serumskih proteina na celogelu (normalni serum (lijevo) i serum sa sporo migrirajućim albuminom Zagreb (desno); 133 kilopiksela; 391 kB pri stupnjevima JPEG-kompresije 3,0 bpp (A1), 0,2 bpp (A2) i 0,1 bpp (A3).

**(B) IZOELEKTRIČNO** fokusiranje na polakrilamidu (normalni albumin bez brzo migrirajućeg albumina Krapina (lijevo) i s njim (desno); 77 kilopiksela; 225 kB pri stupnjevima JPEG-kompresije 3,0 bpp (B1), 0,2 bpp (B2) i 0,1 bpp (B3).

**(C) IZOELEKTRIČNO** fokusiranje na polakrilamidu (otac-normalan albumin, majka i dijete s normalnim i brzo migrirajućim albuminom Krapina; 221 kilopiksela; 649 KB pri stupnjevima JPEG-kompresije 3,0 bpp (C1), 0,2 bpp (C2) i 0,1 bpp (C3). Vrijednost pl od 4,82 predstavlja inačicu albumina, a ostale vrijednosti pripadaju proteinim standardima.

**FIGURE 1.**

**(A) SERUM** protein electrophoresis on cellogel (normal serum (left) and serum containing the slow-migrating albumin Zagreb (right); 133 kilopixels; 391 kB at JPEG-compression degrees of 3.0 bpp (A1), 0.2 bpp (A2) and 0.1 bpp (A3).

**(B) ISOELECTRIC** focusing on polyacrylamide (normal albumin without (left) and with (right) the fast-migrating albumin Krapina; 77 kilopixels; 225 kB at JPEG-compression degrees of 3.0 bpp (B1), 0.2 bpp (B2) and 0.1 bpp (B3).

**(C) ISOELECTRIC** focusing of albumin on polyacrylamide (father-normal, mother and child with both normal albumin and fast-migrating albumin Krapina; 221 kilopixels; 649 kB at JPEG-compression degrees of 3.0 bpp (C1), 0.2 bpp (C2) and 0.1 bpp (C3). The pl-value of 4.82 represents the albumin variant, whereas the remaining values represent protein standards.

najčešće primjenjivana mjerila kvalitete slike (11). PSNR, SNR i MSE predstavljaju realna, objektivna mjerila kvalitete slike (11). Činitelj kvalitete određuje koliko će podataka biti izgubljeno, što izravno odražava veličinu komprimirane slike (12). VCDemo program kompresijskog postupnika JPEG (10) omogućava određivanje OQF, tj. najmanjeg činitelja kvalitete koji daje sliku koja je najsličnija izvornoj slici. SNR podrazumijeva omjer srednje snage signala i efektivne snage šuma, a izračunava se primjenom sljedeće jednadžbe:

$$\text{SNR} = 20 \cdot \log_{10} \left( \frac{A_{\text{signal}}}{A_{\text{noise}}} \right) \quad (1)$$

U toj jednadžbi A predstavlja amplitudu korijena aritmetičke sredine.  $A_{\text{signal}}$  and  $A_{\text{noise}}$  su efektivne vrijednosti amplitude signala, odnosno efektivne vrijednosti amplitude šuma.

MSE je izračunat prema:

$$\text{MSE} = \frac{1}{m} \sum_{i=0}^{m-1} \sum_{j=0}^{n-1} \|I(i, j) - K(i, j)\|^2 \quad (2)$$

gdje je  $I(i, j)$  amplituda piksela izvorne slike, a  $K(i, j)$  amplituda piksela rekonstruirane slike. Parametri m i n predstavljaju predstavljuju broj elemenata slike u horizontalnom i vertikalnom smjeru (izraženo u pikselima).

PSNR je definiran kao:

$$\text{PSNR} = 20 \cdot \log_{10} \left( \frac{\text{MAX}_I}{\sqrt{\text{MSE}}} \right) \quad (3)$$

gdje  $\text{MAX}_I$  predstavlja najveću moguću veličinu elemenata slike, a MSE je srednja kvadratna pogreška (kvadrat zbroja razlike (engl. *Bias*) i varijance procjenitelja). Statistička značajnost testirana je Wilcoxon-Mann-Whitney testom dvostrukog uzorka, a korelacija između dviju varijabli izražena je koeficijentom korelacijske (r). Granična vrijednost OQF, PSNR i MSE određivana je pomoću ROC-analize (13).

## Rezultati

### Subjektivna ocjena

Slike 1.A1, 1.B1, 1.C1 i njihove komprimirane inačice poslane su kao privitak u e-pošti osmorici lokalnih i međunarodnih stručnjaka. Primatelji su se složili da su sve slike komprimirane postupnikom JPEG2000 bile *izvrsne*, tj. bile su jednake kvalitetu kao izvorne slike. S druge strane, kompresija JPEG bila je manje korisna (Tablica 1). Samo sli-

MSE and PSNR are the most often applied measures of image quality (11). PSNR, SNR and MSE are realistic, objective image quality measures (11). The quality factor determines how much data is to be lost, which is directly reflected in the size of a compressed image (12). VCDemo application of the JPEG compression algorithm (10) enables determination of OQF, i.e. the smallest quality factor that yields an image with the highest similarity to the original image. SNR implicates a ratio between medium signal power and effective noise power, and is calculated by applying the following equation:

$$\text{SNR} = 20 \cdot \log_{10} \left( \frac{A_{\text{signal}}}{A_{\text{noise}}} \right) \quad (1)$$

where A represents root mean square amplitude.  $A_{\text{signal}}$  and  $A_{\text{noise}}$  are effective values of signal amplitude and of noise amplitude.

MSE is calculated according to the following equation:

$$\text{MSE} = \frac{1}{m} \sum_{i=0}^{m-1} \sum_{j=0}^{n-1} \|I(i, j) - K(i, j)\|^2 \quad (2)$$

where  $I(i, j)$  is the pixel amplitude of the original image, and  $K(i, j)$  is the pixel amplitude of the reconstructed image. M and n parameters represent the number of elements of an image in horizontal and vertical direction (expressed in pixels).

PSNR is defined as:

$$\text{PSNR} = 20 \cdot \log_{10} \left( \frac{\text{MAX}_I}{\sqrt{\text{MSE}}} \right) \quad (3)$$

where  $\text{MAX}_I$  is the maximum possible pixel value of the image, and MSE is the mean squared error (squared sum of rater bias and variance).

Statistical significance was tested by Wilcoxon-Mann-Whitney two-sample test, and the correlation between two variables was expressed by the coefficient of correlation (r). Cut-off values of OQF, PSNR and MSE were determined using ROC analysis (13).

## Results

### Subjective rating

Figures 1.A1, 1.B1, 1.C1, and their compressed variants were transmitted as email attachments to eight local and international experts. The recipients agreed that all images compressed by JPEG2000 algorithm were *excellent*, i.e. they were of the same quality as original images. On

**TABLICA 1.** Subjektivna ocjena slika pri različitim JPEG-komprezijama**TABLE 1.** Subjective evaluation of images at different JPEG compressions

| Reviewer | Image 1. A1, 1.B1, 1.C1     |                                     |                              |
|----------|-----------------------------|-------------------------------------|------------------------------|
|          | excellent*<br>3.0 - 0.3 bpp | good<br>0.2 bpp                     | useless<br>0.1 bpp           |
| 1        | excellent                   | not so focused but somewhat blurred | useless                      |
| 2        | excellent                   | not so focused but somewhat blurred | useless                      |
| 3        | excellent                   | slightly modified                   | image decomposition          |
| 4        | excellent                   | slightly modified                   | image decomposition          |
| 5        | excellent                   | slightly modified                   | loss of relevant information |
| 6        | excellent                   | slightly modified                   | loss of relevant information |
| 7        | excellent                   | slightly modified                   | image decomposition          |
| 8        | excellent                   | slightly modified                   | image decomposition          |

The same evaluation was obtained for all three images.

\*excellent - the same quality as original image; unchanged; very high resolution.

ke komprimirane stupnjevima 0,3 do 3,0 bpp bile su ocijenjene kao *izvrsne*. Slike komprimirane stupnjem 0,2 bpp ocijenjene su kao *dobre*, a kompresija pri 0,1 bpp rezultirala je *potpuno nekorisnim* slikama. Gubitak kvalitete slika komprimiranih stupnjevima 0,2 i 0,1 bpp postojao je i prije elektronskog prijenosa (uspoređiti slike 2 i 3 sa slikama 1 na Slici 1). prijenos nije utjecao na kvalitetu slika (nije prikazano).

### Objektivna mjerila kvalitete

Tablica 2. prikazuje objektivna mjerila kvalitete 48 slika komprimiranih algoritmima JPEG i JPEG2000. Ponovno, prijenos slika nije utjecao na njihovu kvalitetu. Vrijednosti PSNR nisu se značajno razlikovale između slika kod kompresije  $\geq 0,3$  bpp. Nasuprot tome, postojala je statistički značajna razlika za vrijednosti SNR svake pojedine slike ( $P = 0,029$ ), tj. vrijednost SNR ovisila je o vrsti analizirane slike (sadržaj slike). Međutim, nije postojala statistički značajna razlika između vrijednosti SNR za kompresijske postupnike JPEG i JPEG2000 ( $P = 0,797$ ). Konačno, vrijednosti MSE izračunane za odgovarajuće vrijednosti bpp razlikovale su se značajno ( $P = 0,037$ ) između dviju kompresijskih shema.

Kompresija JPEG kod 0,3 bpp i kompresija JPEG2000 kod 0,1 bpp nisu rezultirale gubitkom relevantnih podataka za uzorke ni elektroforeze ni izoelektričnog fokusiranja, bez obzira na način dobivanja slike. Slike komprimirane postupnikom JPEG, ocijenjene kao *izvrsne*, imale su raspon vrijednosti OQF od 31 do 99 (srednja vrijednost 74; granična vrijednost  $> 20$ ). Postojalo je preklapanje vrijednosti PSNR između slika (kompresija JPEG) ocijenjenih kao *izvrsne* i slika ocijenjenih kao *dobre/nekorisne*. Sve sli-

the other hand, JPEG compression was rated as less useful (Table 1). Only the images compressed at 0.3-3.0 bpp were rated as *excellent*. Compressions at 0.2 bpp were rated as *good*, and compressions at 0.1 bpp resulted in *completely useless* images. Loss in quality of images compressed at 0.2 and 0.1 bpp was present already before electronic transmission (compare images 2 and 3 to images 1 in Fig. 1). The transmission itself did not affect image quality (not shown).

### Objective measures of quality

Table 2 shows objective measures of quality for 48 images compressed by JPEG and JPEG2000 algorithms. Again, image transmission did not have any impact on their quality. PSNR values did not significantly differ between images compressed at  $\geq 0,3$  bpp. In contrast, a statistically significant difference was observed in SNR values for each image ( $P = 0,029$ ), i.e. the SNR value depended on the type of the analyzed image (image content). However, there was no statistically significant difference for SNR values between JPEG and JPEG2000 compression algorithms ( $P = 0,797$ ). Finally, the MSE values calculated for corresponding bpp values significantly differed ( $P = 0,037$ ) between the two compression formats.

JPEG compression at 0.3 bpp and JPEG2000 compression at 0.1 bpp did not result in the loss of relevant data either for electrophoresis or isoelectric focusing samples regardless of the method of image formation. The images compressed by JPEG algorithm and rated as excellent had OQF values ranging from 31 to 99 (mean value 74; cut-off  $> 20$ ). There was an overlap of PSNR values between images (JPEG compression) rated as excellent and images ra-

**TABLICA 2.** Mjerila kvalitete za slike 1.A1, 1.B1 i 1.C1 komprimirane postupnicima JPEG odnosno JPEG2000**TABLE 2.** Quality measures for images 1.A1, 1.B1 i 1.C1 compressed by JPEG or JPEG2000 algorithms

| JPEG                   |                      |           |           |          |           |
|------------------------|----------------------|-----------|-----------|----------|-----------|
| Image                  | Actual bitrate (bpp) | OQF (100) | MSE (1.0) | SNR (dB) | PSNR (dB) |
| GIVEN BITRATE: 3.0 bpp |                      |           |           |          |           |
| A                      | 3.31                 | 99        | 0.3       | 38.6     | 53.1      |
| B                      | 3.03                 | 98        | 0.5       | 29.2     | 50.1      |
| C                      | 2.58                 | 99        | 0.2       | 39.5     | 54.7      |
| GIVEN BITRATE: 1.5 bpp |                      |           |           |          |           |
| A                      | 1.57                 | 94        | 0.9       | 34.1     | 48.7      |
| B                      | 1.49                 | 92        | 1.8       | 24.0     | 45.7      |
| C                      | 1.55                 | 95        | 0.4       | 37.2     | 52.4      |
| GIVEN BITRATE: 1.0 bpp |                      |           |           |          |           |
| A                      | 1.02                 | 87        | 1.4       | 32.0     | 46.5      |
| B                      | 1.01                 | 84        | 2.8       | 22.1     | 43.7      |
| C                      | 0.98                 | 88        | 2.6       | 28.7     | 43.9      |
| GIVEN BITRATE: 0.7 bpp |                      |           |           |          |           |
| A                      | 0.70                 | 66        | 2.2       | 30.2     | 44.8      |
| B                      | 0.69                 | 62        | 4.1       | 20.3     | 42.0      |
| C                      | 0.70                 | 81        | 3.8       | 27.1     | 42.4      |
| GIVEN BITRATE: 0.5 bpp |                      |           |           |          |           |
| A                      | 0.50                 | 61        | 3.3       | 28.4     | 42.9      |
| B                      | 0.49                 | 57        | 5.7       | 18.9     | 40.6      |
| C                      | 0.50                 | 68        | 5.8       | 25.3     | 40.5      |
| GIVEN BITRATE: 0.3 bpp |                      |           |           |          |           |
| A                      | 0.30                 | 31        | 6.5       | 25.5     | 40.0      |
| B                      | 0.30                 | 34        | 8.6       | 17.1     | 38.8      |
| C                      | 0.30                 | 40        | 9.8       | 23.0     | 38.2      |
| GIVEN BITRATE: 0.2 bpp |                      |           |           |          |           |
| A                      | 0.20                 | 12        | 18.1      | 12.0     | 35.5      |
| B                      | 0.20                 | 19        | 13.0      | 15.3     | 37.0      |
| C                      | 0.20                 | 20        | 17.7      | 20.4     | 35.6      |
| GIVEN BITRATE: 0.1 bpp |                      |           |           |          |           |
| A                      | 0.13                 | 2         | 291.0     | 9.0      | 23.5      |
| B                      | 0.14                 | 2         | 297.4     | 1.8      | 23.4      |
| C                      | 0.11                 | 2         | 216.4     | 9.5      | 24.8      |

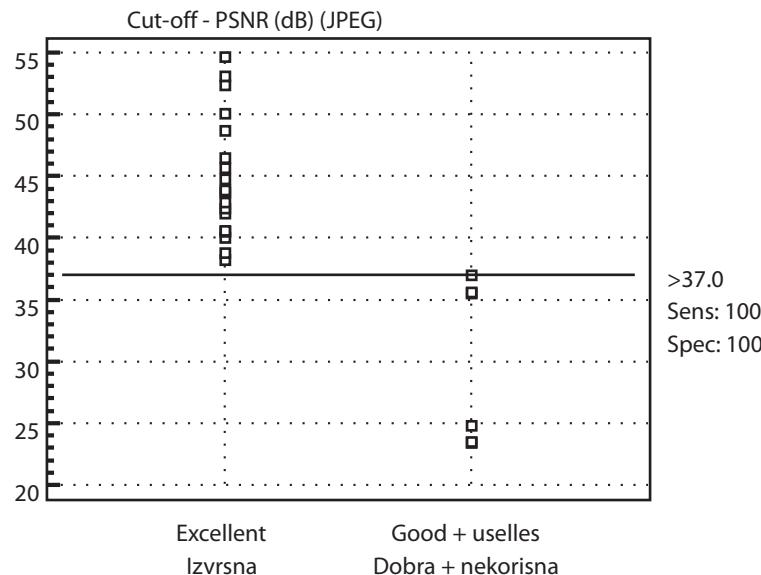
| JPEG2000                |                       |           |          |           |  |
|-------------------------|-----------------------|-----------|----------|-----------|--|
| Actual bitrate (bpp)    | Decoded bitrate (bpp) | MSE (1.0) | SNR (dB) | PSNR (dB) |  |
| GIVEN BITRATE: 3.0 bpp  |                       |           |          |           |  |
| 3.0                     | 1.10                  | 0.7       | 35.4     | 50.0      |  |
| 3.0                     | 1.57                  | 0.7       | 28.0     | 49.7      |  |
| 3.0                     | 1.56                  | 0.5       | 35.5     | 50.7      |  |
| GIVEN BITRATE: 1.5 bpp  |                       |           |          |           |  |
| 1.5                     | 1.10                  | 0.7       | 35.4     | 50.0      |  |
| 1.5                     | 1.50                  | 0.8       | 27.7     | 49.3      |  |
| 1.5                     | 1.50                  | 0.6       | 35.1     | 50.4      |  |
| GIVEN BITRATE: 1.0 bpp  |                       |           |          |           |  |
| 1.0                     | 1.00                  | 0.8       | 34.8     | 49.4      |  |
| 1.0                     | 1.00                  | 1.6       | 24.4     | 46.0      |  |
| 1.0                     | 1.00                  | 1.5       | 31.2     | 46.4      |  |
| GIVEN BITRATE: 0.75 bpp |                       |           |          |           |  |
| 0.75                    | 0.75                  | 1.1       | 33.1     | 47.7      |  |
| 0.75                    | 0.75                  | 2.3       | 22.8     | 44.4      |  |
| 0.75                    | 0.75                  | 2.2       | 29.5     | 44.7      |  |
| GIVEN BITRATE: 0.5 bpp  |                       |           |          |           |  |
| 0.50                    | 0.50                  | 1.7       | 31.2     | 45.7      |  |
| 0.50                    | 0.50                  | 3.8       | 20.7     | 42.4      |  |
| 0.50                    | 0.50                  | 3.5       | 27.5     | 42.7      |  |
| GIVEN BITRATE: 0.3 bpp  |                       |           |          |           |  |
| 0.30                    | 0.30                  | 2.9       | 29.0     | 43.5      |  |
| 0.30                    | 0.30                  | 5.5       | 19.1     | 40.7      |  |
| 0.30                    | 0.30                  | 5.4       | 25.5     | 40.8      |  |
| GIVEN BITRATE: 0.2 bpp  |                       |           |          |           |  |
| 0.20                    | 0.20                  | 4.0       | 27.6     | 42.2      |  |
| 0.20                    | 0.20                  | 7.1       | 18.0     | 39.6      |  |
| 0.20                    | 0.20                  | 7.8       | 24.0     | 39.2      |  |
| GIVEN BITRATE: 0.1 bpp  |                       |           |          |           |  |
| 0.10                    | 0.10                  | 6.6       | 25.4     | 39.9      |  |
| 0.10                    | 0.10                  | 9.7       | 16.6     | 38.3      |  |
| 0.10                    | 0.10                  | 15.0      | 21.1     | 36.4      |  |

ke prihvaćene kao izvrsne imale su vrijednosti PSNR > 37 dB (granična vrijednost) (Slika 2). Vrijednosti PSNR (srednje vrijednosti) za slike A1, B1 i C1 su u skladu sa vrijednostima za slike komprimirane postupnicima JPEG2000.

ted as *good/useless*. All images received as excellent had PSNR values > 37 dB (cut-off value) (Fig. 2). The values of

**SLIKA 2.** Granična vrijednost za PSNR kod kompresije JPEG. Sve slike prihvaćene kao *izvrsne* imale su vrijednost PSNR > 37dB (osjetljivost 100%), a sve slike prihvaćene kao *dobre ili nekorisne* imale su vrijednost PSNR ≤ 37dB (specifičnost 100%).

**FIGURE 2.** Cut-off value for PSNR at JPEG compression. All images accepted as *excellent* had a PSNR-value > 37dB (sensitivity 100%), while all images accepted as *good or useless* had a PSNR-value ≤ 37dB (specificity 100%).



Sens: 100 - Sensitivity 100% = 100% probability that PSNR will be > 37dB when images are evaluated as *excellent*.  
 Spec: 100 - Specificity 100% = 100% probability that PSNR will be ≤ 37dB when images are evaluated as *good or useless*.

ja vrijednost 39,0 dB) i SNR (srednja vrijednost 21,9 dB) pri kompresiji od 0,3 bpp za kompresiju JPEG mogle su se usporediti s vrijednostima PSNR (srednja vrijednost 38,2 dB) i SNR (srednja vrijednost 21,0 dB) dobivenih kod 0,1 bpp pri kompresiji JPEG2000. Granične vrijednosti MSE za slike komprimirane postupnikom JPEG, ocijenjene kao izvrsne, bile su ≤ 9,8. Pri istim dvjema vrijednostima bpp srednja vrijednost za MSE bila je 8,3 za JPEG, odnosno 10,4 za JPEG2000.

Pri kompresiji JPEG, PSNR i SNR su bili u pozitivnoj korelaciji s OQF, a negativnoj korelaciji s MSE (Tablica 3.). Korelacija između OQF i MSE bila je negativna. PSNR je imao pozitivnu korelaciju s aktualnom kompresijom za JPEG i za JPEG2000. Jednako tako, pri kompresiji JPEG2000, PSNR i MSE kao i SNR i MSE pokazali su međusobnu negativnu korelaciju. Sve te negativne korelacije utemeljene su na teoriji, jer je MSE mjerilo pogreške, a PSNR i SNR su mjerila kvalitete (jednadžba 3).

## Rasprava

Subjektivnim ocjenjivanjem sve su slike komprimirane algoritmom JPEG2000 ocijenjene kao *izvrsne*. Protivno tome, kod kompresije JPEG stupnjem 0,1 bpp slike su ocijenjene kao *potpuno neupotrebljive*, kompresijom od 0,2 bpp ocijenjene su kao *dobre ili umjereni nejasne*, a slike

PSNR (mean value 39.0 dB) and SNR (mean value 21.9 dB) at JPEG compression at 0.3 bpp could be compared to the values of PSNR (mean value 38.2 dB) and SNR (mean value 21.0 dB) obtained at JPEG2000 compression at 0.1 bpp. MSE cut-off values for images compressed by JPEG algorithm that were rated as excellent were ≤ 9.8. At the same bpp values, mean MSE values were 8.3 and 10.4 for JPEG and JPEG2000, respectively.

In JPEG compression, both PSNR and SNR correlated positively to OQF, and negatively to MSE (Table 3). PSNR was in positive correlation to the actual JPEG and JPEG2000 compressions. In JPEG2000 compression, PSNR and MSE, and SNR and MSE showed mutually negative correlation. All these negative correlations are theoretically based since MSE is an error measure, while PSNR and SNR are measures of quality (Equation 3).

## Discussion

All images compressed by JPEG2000 algorithm were subjectively rated as excellent. In contrast, JPEG compression at 0.1 bpp resulted in images rated as completely useless; images compressed at 0.2 bpp were rated as good or moderately blurred, while those compressed at 0.3-3.0 bpp were rated as excellent. Higher degrees of compression with less distortion can be achieved using JPEG2000 algo-

**TABLICA 3.** Koeficijenti korelacije (r) za objektivna mjerila kvalitete slika komprimiranih postupnicima JPEG, odnosno JPEG2000**TABLE 3.** Correlation coefficients (r) for objective quality measures of images compressed by JPEG and JPEG2000 algorithms

|                      | r     | P      |
|----------------------|-------|--------|
| <b>JPEG</b>          |       |        |
| PSNR: OQF            | 0,92  | <0.001 |
| SNR: OQF             | 0,88  | <0.001 |
| PSNR: MSE            | -0,81 | <0.001 |
| SNR: MSE             | -0,71 | <0.001 |
| OQF: MSE             | -0,65 | <0.001 |
| PSNR: actual bitrate | 0,78  | <0.001 |
| <b>JPEG2000</b>      |       |        |
| PSNR: MSE            | -0,90 | <0.001 |
| SNR: MSE             | -0,73 | <0.001 |
| PSNR: actual bitrate | 0,81  | <0.001 |

komprimirane od 0,3 do 3,0 bpp ocijenjene su kao *izvrsne*. Algoritmom JPEG2000 mogu se postići veći stupnjevi kompresije s manje izobličenja nego, primjerice, postupnikom JPEG (14). S kliničkog stajališta, subjektivno vrednovanje kvalitete slika od prvotnog je značenja. Glede objektivnog ocjenjivanja kvalitete slike, nije lako pronaći objektivna brojčana mjerila koja bi bila korisna za sve kompresijske metode (7).

PSNR, SNR i MSE su objektivna mjerila kvalitete slike koja se najčešće primjenjuju (11). U ovom radu je postojala statistički značajna razlika između vrijednosti MSE određenih za pojedine slike i vrijednosti MSE izračunane za dvije kompresijske sheme. Prema Veldhuizenu (15), MSE ovisi o mjerenu intenziteta slike. Vrijednost PSNR različitih slika nisu se značajno razlikovale. Budući da PSNR ne ovisi o energiji signala (za razliku od SNR, koji o njoj ovisi), on ukazuje na količinu (energije) šuma koji utječe na sliku. Budući da je MSE mjerilo pogreške, a PSNR i SNR mjerila kvalitete, njihov međusoban odnos je aproksimativno obratno razmjeran, pa su vrijednosti MSE bile u negativnoj korelaciji s PSNR i SNR. Stoga su SNR vrijednosti ovisile o vrsti analizirane slike. Međutim, nije postojala statistički značajna razlika između vrijednosti SNR dobivenih za dvije kompresijske sheme. Zanimljivo je da su naša prijašnja ispitivanja kvalitete slika radiograma pokazala da su samo vrijednosti PSNR u digitaliziranim slikama bile veće nego srednje vrijednosti digitalnih fotografija, kako kod JPEG tako i kod JPEG2000 (16). Ovdje smo uočili da kompresija JPEG kod 0,3 bpp i kompresija JPEG2000 kod 0,1 bpp nisu rezultirale gubitkom relevantnih podataka ni kod elektroforeze ni kod izoelektričnog fokusiranja, bez obzira na način dobivanja slike. Važno je naglasiti da

rithm than by, e.g., using JPEG algorithm. Clinically, subjective evaluation of image quality is of primary significance. Regarding objective evaluation of image quality, it is not easy to find objective numerical measures that would be useful for all compression methods (7).

PSNR, SNR and MSE are objective measures of image quality that are applied most frequently (11). In this study, a statistically significant difference was observed between MSE values determined for some images and MSE values calculated for the two compression schemes. According to Veldhuizen (15), MSE depends on the scaling of image intensity. PSNR values of different images did not differ significantly. As PSNR does not depend on signal power (unlike SNR), it shows the amount (power) of noise that affects an image. Also, since MSE is an error measure while PSNR and SNR are quality measures, their interrelationship was approximately inversely proportional so that MSE values were negatively correlated to PSNR and SNR. Therefore, SNR values were dependent on the type of the analyzed image. However, no statistically significant difference was observed between the SNR values obtained for the two compression schemes. It is interesting that our previous investigations of the quality of x-ray images demonstrated that only PSNR values for digitalized images were higher than mean values of digital photos, both for JPEG and JPEG2000 (16). In this study, we noted that JPEG compression at 0.3 bpp and JPEG2000 compression at 0.1 bpp did not result in the loss of relevant data either in electrophoresis or isoelectric focusing, regardless of how an image was obtained. It is important to point out that PSNR and SNR values (39.0 and 21.9 dB, respectively) at the JPEG compression rate of 0.3 bpp corresponded to

vrijednosti PSNR i SNR (39,0 odnosno 21,9 dB) pri stupnju kompresije JPEG od 0,3 bpp odgovaraju PSNR i SNR vrijednostima (38,2 odnosno 21,0 dB) pri stupnju kompresije JPEG2000 od 1,0 bpp. Odgovarajuća vrijednost MSE iznosila je 8,3 za JPEG odnosno 10,4 za JPEG2000. Sve slike prihvaćene kao *izvrsne* imale su vrijednost PSNR > 37 dB, a sve slike prihvaćene kao *dobre* ili *nekorisne* imale su vrijednost PSNR ≤ 37dB (Slika 2). Ta se granična vrijednost PSNR može primijeniti za razlikovanje slika velike kvalitete od slika slabe kvalitete. Ipak, granične vrijednosti PSNR su, prema Ashraf i sur. (17), različite za različite slike (slika mrežnice - 30,2dB; angiogram - 38,3 dB; radiogram pluća - 31,2 dB; sve vrijednosti za algoritam JPEG) (17). Yamamoto i sur. (18) pokazali su da su vrijednosti PSNR za slike CT pluća velike kvalitete bile u rasponu od 45,3 do 44,1 dB. U ranijem istraživanju, granične vrijednosti PSNR za digitalizirane plućne radiograme iznosile su 45,3 dB (za JPEG pri 0,3 bpp), odnosno 44,1 dB (za JPEG2000 pri 0,1 bpp) (17,19). Prema tome, čini se da vrijednosti PSNR ovise o sadržaju slike. Zapravo, ono što ovisi o sadržaju slike jest mogućnost odgovarajuće metode kompresije: slika s manje detalja lakše se komprimira nego slika s mnogo detalja. Chandra i sur. su pokazali da je QF dobar pokazatelj kvalitete slika komprimiranih algoritmom JPEG (20), te da OQF od 75 podrazumijeva slike *izvrsne* kvalitete, što je u skladu s našim radom, koji je dao srednje vrijednosti za OQF 74. OQF je korelirao pozitivno s PSNR i SNR, a negativno s MSE. Iz rezultata u tablici 2. vidi se da iste vrijednosti *bit rate* rezultiraju različitim OQF vrijednostima (kod *bit rate* 1,0, 0,7, 0,5, 0,3 i 0,2), što je u skladu s rezultatima Fidlera i sur. (21) koji su pokazali da je vrijednosti OQF važna za reproducibilnost detalja slike. Jedno od ograničenja našeg istraživanja svakako je bio mali broj izvornih slika, što je moglo utjecati na rezultate statističke analize. U mogućem budućem istraživanju trebalo bi analizirati veći broj izvornih slika različitog sadržaja te odabrat i neka druga mjerila objektivne kvalitete slike. Dodatno ograničenje jest u činjenici da su procjenitelji kvalitete znali (engl. *not blinded*) koji su elementi važni za procjenu vrste slike, što je moglo utjecati na njihovu subjektivnu procjenu. Prednost treba dati kompresijskom postupniku JPEG2000, što je i preporuka *National Electrical Manufacturers Association*, koja je standard JPEG2000 pridodata standardima DICOM (engl. *Digital Imaging and Communications in Medicine*) za kompresiju medicinskih slika (22). Ako se primjenjuje standard JPEG, treba voditi računa o stupnju kompresije do kojeg se slika može komprimirati, a da joj kvaliteta bude očuvana. U Hrvatskoj je zanimanje za telemedicinu usmjereno prema virtualnim e-medicinskim centrima (23), e-domovima zdravlja i otočnoj medicini (24), te telekirurgiji (25). Preporuke za *online-komunikaciju* u telehematologiji i ostalim telemedicinskim konzultacijama dostupne su u Pravilniku o obavljanju telekomunikacijskih konzultacija (samo hrvatska inačica) (26). Budući da se međunarodna jednoznačnost metoda u telemedicini

PSNR and SNR values (38.2 and 21.0 dB, respectively) obtained for JPEG2000 compression at 1.0 bpp. The corresponding MSE value was 8.3 for JPEG and 10.4 for JPEG2000. All images rated as excellent had the PSNR value > 37 dB, and all images received as good or useless had PSNR value ≤ 37dB (Fig. 2). This PSNR cut-off value may be applied to distinguish high quality- from low quality images. Still, PSNR cut-off values are, according to Ashraf et al. (17) different for different images (retinal image: 30.2 dB; angiogram: 38.3 dB; lung x-ray: 31.2 dB, all for JPEG algorithm) (17). Yamamoto et al. (18) showed that PSNR values for high quality CT lung images ranged from 45.3 to 44.1 dB. In a previous study, PSNR cut-off values for digitalized lung x-rays were 45.3 dB for JPEG at 0.3 bpp and 44.1 dB for JPEG2000 at 0.1 bpp (17,19). Accordingly, it appears that PSNR values depend on image content. Actually, the capacity of an adequate compression method is related to image content: an image with less details is easier to compress than an image with many details. Chandra et al. demonstrated QF to be a good indicator of the quality of images compressed by JPEG algorithm (20), and showed that OQF value of 75 stands for images of excellent quality, which is consistent with our study where mean OQF value was 74. OQF correlated positively with PSNR and SNR, and negatively with MSE. The results presented in Table 2 reveal that equal bit rate values yielded different OQF values (at 1.0, 0.7, 0.5, 0.3 and 0.2 bit rates), which is consistent with results by Fidler et al. (21) who showed the OQF value to be important for reproducibility of image details. One of the limitations of our study was certainly a low number of original images, which might have influenced statistical analysis results. In a potential future investigation it would be necessary to analyze a higher number of original images with different content and also choose some other objective measures to establish image quality. Additional limitation is the fact that quality assessors were not blinded to the elements that were important for assessing image type, which might have resulted in a bias in their subjective rating.

Precedence should be given to JPEG2000 compression algorithm, which was actually recommended by the National Electrical Manufacturers Association that included JPEG2000 among DICOM (Digital Imaging and Communications in Medicine) standards for compression of medical images (22). If JPEG standard is applied, care should be taken of the highest degree of compression that can be used to maintain image quality at the same time. In Croatia, interest in telemedicine is focused on virtual e-medical centers (23), e-health care centers and island medicine (24), and telesurgery (25). Recommendations for online communication in telehematology and other telemedical consultations are available in the Rules on Implementing Telecommunication Consultations (only in Croatian) (26). As international uniformity of methods in telemedicine progresses, we suggest that, except in telemicroscopy,

ni unapređuje, predlažemo da se osim u telemikroskopiji kompresijski postupnik JPEG2000, nakon ocjene kvalitete većeg broja slika različitog sadržaja, primjeni i za slikovne zapise dobivene u medicinsko-biokemijskom laboratoriju.

## Zahvala

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## Adresa za dopisivanje:

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JPEG compression algorithm is, after assessment of a substantial number of images of different content, also used for image files produced in a medical biochemistry laboratory.

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