

Digital Image Basics

Pixel and voxel concepts

2D picture elements, 3D volume elements

Bit depth

8-bit (256 levels), 16-bit (65,536 levels)

File formats

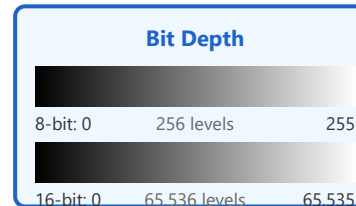
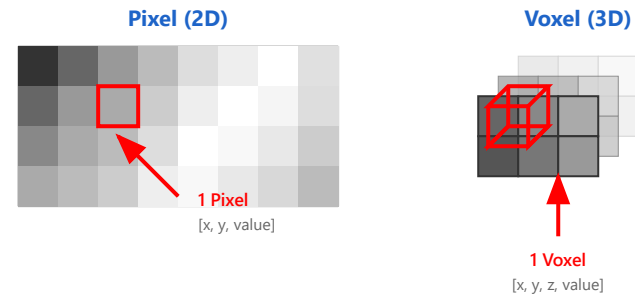
TIFF, PNG (lossless), JPEG (lossy)

Compression methods

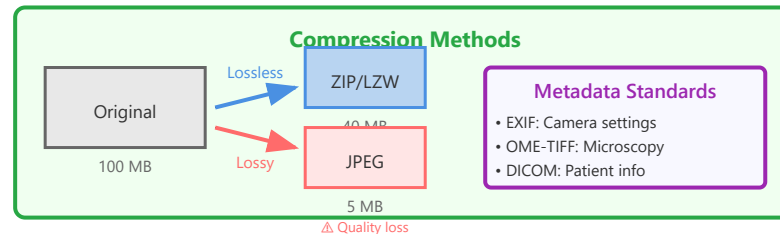
Lossless vs lossy tradeoffs

Metadata standards

EXIF, OME-TIFF for scientific imaging



File Format Comparison		
Format	Compression	Use Case
TIFF	Lossless	Scientific
PNG	Lossless	Web/Analysis
JPEG	Lossy	Web/Display
DICOM	Both	Medical



1. Pixel and Voxel Concepts

Pixels: The Building Blocks of 2D Images

A pixel (picture element) is the smallest addressable element in a digital image. Each pixel contains intensity information that represents color or brightness at a specific location.

- **Coordinates:** $[x, y]$ position in the image grid
- **Value:** Intensity level (e.g., 0-255 for 8-bit)
- **Color:** May contain RGB channels (Red, Green, Blue)
- **Size:** Determined by image resolution (e.g., 1920×1080)

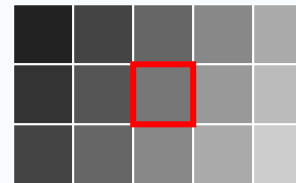
Voxels: 3D Extension of Pixels

A voxel (volume element) represents a value in 3D space, commonly used in medical imaging (CT, MRI) and microscopy.

- **Coordinates:** $[x, y, z]$ position in 3D space
- **Applications:** Medical imaging, 3D microscopy, scientific visualization
- **Volume:** Defined by spacing in x, y, z dimensions

Key Difference: Pixels are 2D (flat images), while voxels add depth information for 3D visualization and analysis.

2D Pixel Structure



Selected Pixel Info

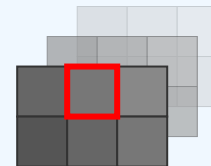
Position: $[3, 2]$

Intensity: 119

8-bit value

Grayscale Display

3D Voxel Structure



Selected Voxel Info

Position: $[2, 1, 2]$

Intensity: 119

Slice: $Z=2$

Common in: MRI, CT, μ -CT

2. Bit Depth

Understanding Bit Depth

Bit depth determines the number of possible intensity values per pixel. Higher bit depth allows for more subtle differences in brightness and color.

- **8-bit:** $2^8 = 256$ levels (0-255)
- **16-bit:** $2^{16} = 65,536$ levels (0-65,535)
- **32-bit:** Used for HDR and scientific data

Practical Applications

8-bit Images: Sufficient for most display purposes, web graphics, and standard photography. File sizes are smaller and processing is faster.

16-bit Images: Essential for scientific imaging, astronomy, microscopy, and professional photography where subtle intensity differences matter.

Dynamic Range Impact

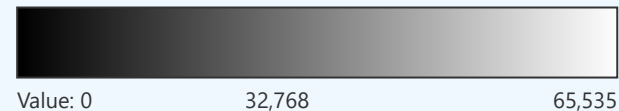
- **Banding:** 8-bit may show visible steps in smooth gradients
- **Detail:** 16-bit captures fine intensity variations
- **Post-processing:** Higher bit depth preserves quality during editing

8-bit (256 levels)



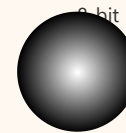
Standard for web images and displays

16-bit (65,536 levels)



Scientific imaging and professional photography

Visual Comparison



Visible steps



Smooth gradient

Memory
8-bit: 1 MB
16-bit: 2 MB

Trade-off: Higher bit depth = Better quality but larger file sizes and slower processing

3. File Formats

Lossless Formats

TIFF (Tagged Image File Format):

- Supports 8-bit to 32-bit images
- Can store multiple pages/layers
- Ideal for archival and scientific data
- Large file sizes but perfect quality

PNG (Portable Network Graphics):

- Supports transparency (alpha channel)
- Efficient compression without quality loss
- Web-friendly, widely supported
- Good for graphics with sharp edges

Lossy Format

JPEG (Joint Photographic Experts Group):

- Aggressive compression for small file sizes
- Quality degrades with each save
- Best for photographs and natural images
- Adjustable quality/compression ratio
- No transparency support

Choosing the Right Format: Use TIFF for scientific work, PNG for web graphics with transparency, JPEG for photographs where some quality loss is acceptable.

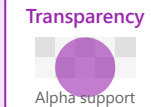
TIFF

Compression: Lossless (LZW, ZIP) or None
Bit Depth: 8, 16, 32-bit support
Use Case: Scientific imaging, archival

File Size
Large
~5-50 MB

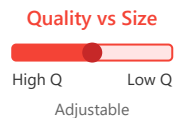
PNG

Compression: Lossless (DEFLATE)
Bit Depth: 8, 16-bit, alpha channel
Use Case: Web graphics, screenshots



JPEG

Compression: Lossy (DCT-based)
Bit Depth: 8-bit only
Use Case: Photography, web display



Format Selection Guide

✓ Scientific Data	→ TIFF, HDF5
✓ Web Graphics	→ PNG, WebP
✓ Photography	→ JPEG, HEIF

4. Compression Methods

Lossless Compression

Preserves all original data - perfect reconstruction is possible.

- **ZIP/DEFLATE:** Used in PNG, general-purpose compression
- **LZW:** Used in TIFF and GIF files
- **Run-Length Encoding (RLE):** Efficient for images with large uniform areas
- **Huffman Coding:** Variable-length encoding based on frequency

Compression ratio: Typically 2:1 to 4:1, depending on image content

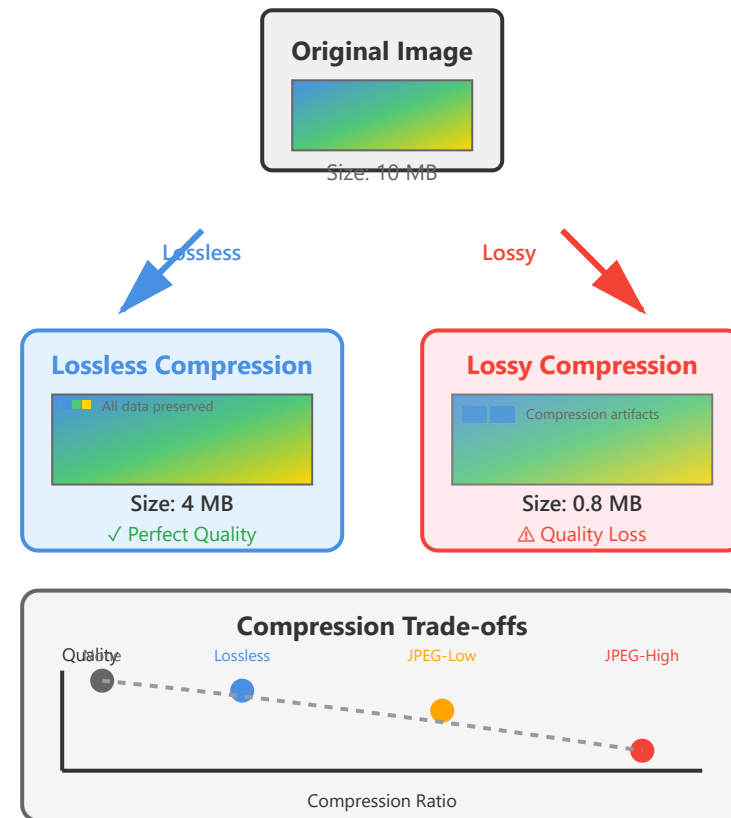
Lossy Compression

Discards some data to achieve higher compression ratios.

- **JPEG (DCT):** Discrete Cosine Transform, exploits human vision limitations
- **JPEG 2000:** Wavelet-based, better quality at high compression
- **WebP:** Modern format combining best of both approaches

Compression ratio: 10:1 to 50:1 or higher, with visible quality degradation

Key Consideration: Lossy compression is irreversible - save originals before compressing for distribution.



5. Metadata Standards

EXIF (Exchangeable Image File Format)

Standard metadata format embedded in JPEG, TIFF, and other image files.

- **Camera Settings:** ISO, aperture, shutter speed, focal length
- **Date & Time:** When the image was captured
- **Location:** GPS coordinates (if available)
- **Device Info:** Camera model, lens information
- **Image Properties:** Resolution, orientation, color space

OME-TIFF (Open Microscopy Environment)

Specialized format for microscopy and scientific imaging.

- **Multi-dimensional data:** X, Y, Z, time, channels
- **Acquisition details:** Microscope settings, objectives
- **Physical dimensions:** Pixel size in micrometers
- **Channel information:** Fluorophore names, wavelengths
- **Experimental metadata:** Sample info, protocols

DICOM (Medical Imaging)

- Patient information and medical history
- Scan parameters and protocols
- Institutional data for regulatory compliance

EXIF Metadata



Camera Information:

- Make: Canon EOS R5
- Lens: RF 24-105mm f/4
- ISO: 400
- Exposure: 1/250s at f/5.6
- Date: 2025-11-19 14:30:22
- GPS: 37.5665°N, 126.9780°E

OME-TIFF Metadata



Microscopy Metadata:

- Dimensions: 512×512×50 (X×Y×Z)
- Pixel Size: 0.65 µm/pixel
- Channels: DAPI, GFP, mCherry
- Objective: 40× NA 1.3
- Time Points: 100 frames
- Exposure: 100ms per channel

DICOM Metadata



Medical Imaging Metadata:

- Patient ID: [Protected]
- Study Date: 2025-11-15
- Modality: MRI / CT / X-Ray
- Body Part: Brain / Chest / Abdomen
- Institution: Medical Center XYZ
- Slice Thickness: 1.0 mm

Importance: Metadata ensures reproducibility, enables batch analysis, and preserves essential information about image acquisition.