

# IMAGE PROCESSING

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## AN INTRODUCTION

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# WHAT IS AN IMAGE?

- ▶ Just a bunch of numbers
- ▶ Organized in a 2D matrix
- ▶ The value of which are mapped to an intensity or color

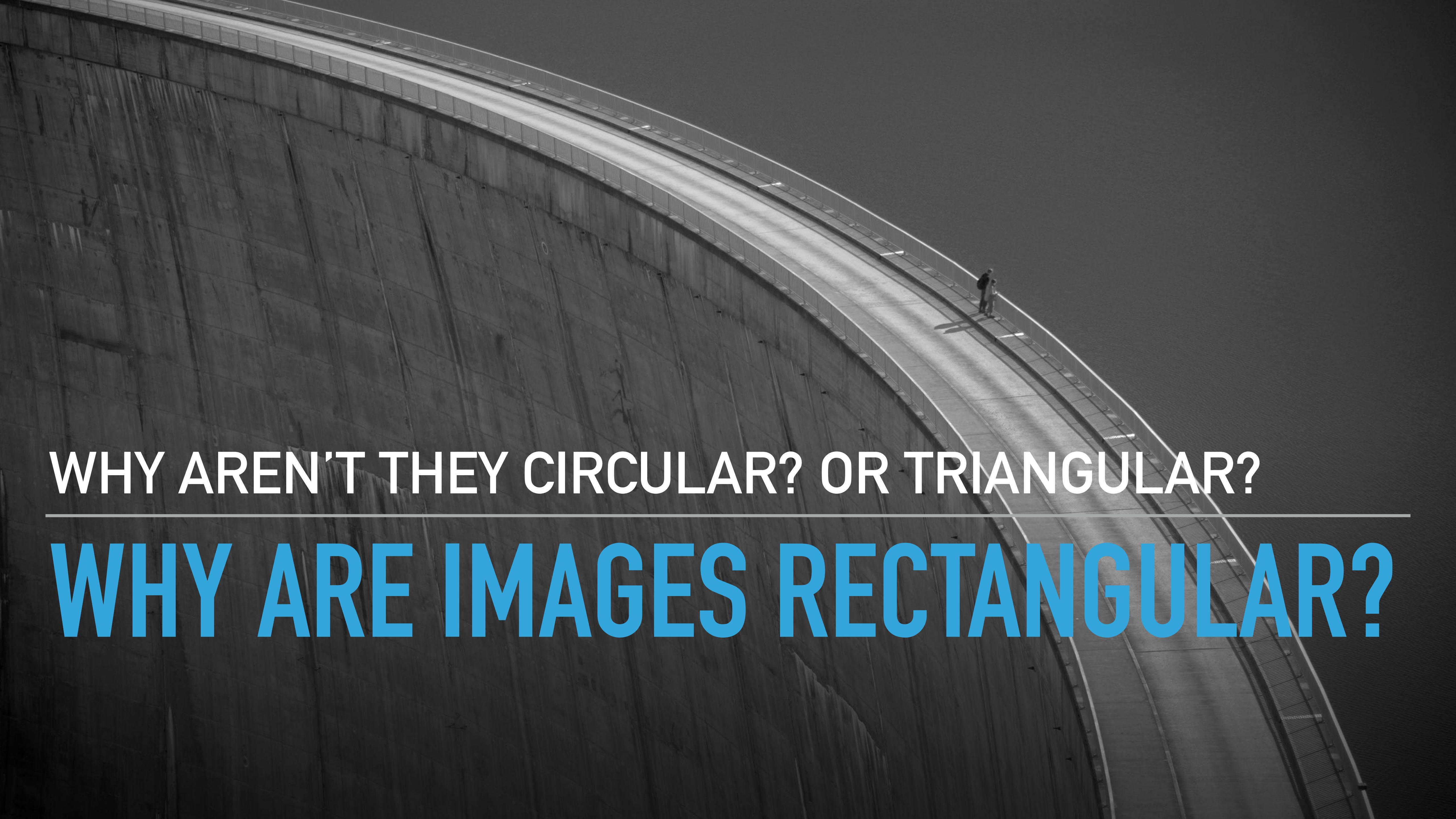




|     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| 220 | 220 | 215 | 203 | 200 | 184 | 178 | 178 | 178 | 179 | 185 | 188 | 188 | 187 | 182 | 180 | 173 | 166 | 163 | 156 | 155 | 158 | 159 | 166 | 172 | 175 | 187 | 190 | 193 | 198 | 198 | 193 | 167 | 157 | 141 | 127 | 121 | 94  | 90  | 86  | 81  | 85  |     |     |
| 243 | 244 | 240 | 233 | 229 | 213 | 207 | 204 | 192 | 191 | 187 | 185 | 182 | 179 | 178 | 170 | 169 | 164 | 160 | 157 | 152 | 150 | 148 | 148 | 153 | 159 | 162 | 176 | 180 | 187 | 196 | 199 | 207 | 206 | 190 | 184 | 167 | 152 | 146 | 121 | 117 | 115 | 114 | 120 |
| 252 | 253 | 250 | 244 | 241 | 224 | 218 | 213 | 198 | 195 | 187 | 183 | 179 | 175 | 173 | 162 | 160 | 158 | 156 | 153 | 148 | 146 | 143 | 142 | 146 | 150 | 154 | 169 | 174 | 183 | 193 | 197 | 209 | 209 | 199 | 196 | 179 | 165 | 159 | 135 | 131 | 130 | 130 | 136 |
| 247 | 248 | 246 | 241 | 238 | 222 | 216 | 211 | 197 | 194 | 182 | 177 | 169 | 162 | 160 | 153 | 152 | 151 | 149 | 146 | 139 | 137 | 133 | 131 | 135 | 139 | 141 | 155 | 159 | 167 | 177 | 182 | 194 | 196 | 199 | 199 | 188 | 178 | 174 | 157 | 154 | 153 | 151 | 155 |
| 241 | 243 | 242 | 238 | 236 | 221 | 215 | 210 | 195 | 191 | 177 | 171 | 160 | 150 | 148 | 143 | 142 | 142 | 141 | 138 | 130 | 128 | 122 | 120 | 123 | 126 | 128 | 140 | 144 | 152 | 162 | 167 | 178 | 183 | 197 | 202 | 196 | 191 | 189 | 179 | 177 | 175 | 172 | 172 |
| 230 | 232 | 230 | 226 | 223 | 211 | 206 | 201 | 188 | 184 | 169 | 163 | 151 | 141 | 138 | 135 | 134 | 135 | 136 | 133 | 127 | 124 | 118 | 116 | 118 | 121 | 122 | 134 | 136 | 143 | 152 | 156 | 168 | 173 | 189 | 195 | 191 | 188 | 186 | 177 | 175 | 170 | 163 | 162 |
| 201 | 203 | 202 | 199 | 197 | 187 | 183 | 180 | 169 | 165 | 150 | 144 | 131 | 121 | 119 | 119 | 122 | 126 | 124 | 120 | 118 | 112 | 110 | 113 | 115 | 117 | 123 | 125 | 129 | 133 | 137 | 146 | 152 | 171 | 178 | 178 | 178 | 176 | 164 | 160 | 148 | 134 | 128 |     |
| 194 | 195 | 195 | 192 | 191 | 182 | 178 | 174 | 164 | 160 | 145 | 140 | 127 | 116 | 115 | 116 | 121 | 125 | 123 | 119 | 117 | 113 | 111 | 113 | 114 | 116 | 121 | 123 | 125 | 128 | 131 | 140 | 145 | 163 | 170 | 170 | 171 | 169 | 158 | 154 | 141 | 125 | 118 |     |
| 177 | 177 | 179 | 183 | 182 | 177 | 175 | 171 | 160 | 157 | 144 | 140 | 130 | 122 | 120 | 117 | 117 | 125 | 133 | 132 | 131 | 127 | 125 | 124 | 122 | 121 | 117 | 115 | 114 | 111 | 113 | 118 | 120 | 121 | 122 | 123 | 124 | 123 | 114 | 111 | 100 | 87  | 82  |     |
| 170 | 171 | 174 | 179 | 179 | 176 | 174 | 170 | 158 | 155 | 144 | 139 | 132 | 125 | 123 | 116 | 125 | 135 | 136 | 136 | 136 | 132 | 130 | 127 | 126 | 124 | 115 | 113 | 110 | 105 | 106 | 110 | 110 | 106 | 105 | 106 | 107 | 106 | 98  | 95  | 85  | 73  | 68  |     |
| 174 | 174 | 175 | 177 | 177 | 173 | 172 | 168 | 159 | 157 | 147 | 144 | 138 | 134 | 132 | 126 | 126 | 133 | 142 | 142 | 143 | 142 | 137 | 135 | 131 | 128 | 126 | 115 | 113 | 108 | 102 | 102 | 101 | 96  | 94  | 94  | 93  | 86  | 84  | 76  | 68  | 64  |     |     |
| 181 | 179 | 178 | 176 | 175 | 168 | 166 | 164 | 160 | 159 | 157 | 157 | 157 | 153 | 153 | 156 | 159 | 159 | 159 | 158 | 150 | 148 | 139 | 132 | 129 | 115 | 111 | 103 | 94  | 91  | 81  | 78  | 68  | 64  | 60  | 58  | 55  | 54  | 55  | 56  | 55  | 55  |     |     |
| 182 | 180 | 178 | 175 | 174 | 168 | 166 | 165 | 160 | 159 | 160 | 159 | 159 | 158 | 158 | 159 | 162 | 162 | 161 | 159 | 154 | 151 | 142 | 134 | 131 | 115 | 111 | 102 | 92  | 88  | 78  | 74  | 65  | 61  | 58  | 55  | 52  | 52  | 54  | 55  | 55  | 55  |     |     |
| 180 | 177 | 175 | 175 | 170 | 169 | 164 | 162 | 162 | 160 | 162 | 162 | 163 | 165 | 165 | 165 | 166 | 166 | 164 | 163 | 159 | 157 | 149 | 142 | 138 | 117 | 112 | 100 | 85  | 80  | 67  | 64  | 59  | 57  | 56  | 55  | 56  | 55  | 55  | 55  | 55  | 55  |     |     |
| 177 | 175 | 172 | 167 | 165 | 161 | 159 | 159 | 158 | 159 | 161 | 162 | 163 | 165 | 166 | 167 | 168 | 168 | 167 | 164 | 164 | 161 | 145 | 140 | 116 | 111 | 97  | 81  | 76  | 64  | 60  | 57  | 55  | 56  | 57  | 57  | 55  | 54  | 53  |     |     |     |     |     |
| 165 | 163 | 161 | 155 | 154 | 150 | 149 | 149 | 151 | 152 | 155 | 157 | 159 | 159 | 159 | 160 | 159 | 158 | 157 | 158 | 159 | 160 | 160 | 151 | 144 | 138 | 112 | 106 | 91  | 74  | 69  | 58  | 55  | 50  | 49  | 52  | 54  | 54  | 55  | 52  | 50  | 49  |     |     |
| 154 | 152 | 149 | 145 | 143 | 140 | 139 | 141 | 144 | 145 | 150 | 151 | 153 | 155 | 154 | 150 | 149 | 148 | 148 | 150 | 155 | 157 | 159 | 160 | 150 | 143 | 137 | 108 | 102 | 86  | 67  | 63  | 51  | 48  | 45  | 43  | 47  | 50  | 51  | 53  | 52  | 49  | 46  | 45  |
| 146 | 144 | 141 | 137 | 135 | 132 | 132 | 133 | 138 | 139 | 142 | 144 | 145 | 147 | 146 | 141 | 140 | 141 | 141 | 143 | 150 | 152 | 153 | 154 | 143 | 134 | 128 | 98  | 91  | 75  | 58  | 55  | 46  | 44  | 41  | 40  | 44  | 48  | 49  | 50  | 46  | 42  | 41  |     |
| 129 | 129 | 127 | 123 | 122 | 120 | 119 | 120 | 122 | 123 | 126 | 128 | 129 | 128 | 128 | 124 | 123 | 124 | 126 | 129 | 136 | 138 | 140 | 141 | 127 | 113 | 106 | 73  | 65  | 52  | 38  | 37  | 33  | 35  | 36  | 38  | 41  | 41  | 43  | 43  | 39  | 36  | 35  |     |
| 125 | 126 | 124 | 119 | 118 | 116 | 115 | 116 | 119 | 120 | 122 | 124 | 123 | 123 | 122 | 118 | 117 | 121 | 124 | 133 | 135 | 136 | 121 | 108 | 100 | 67  | 60  | 47  | 33  | 32  | 32  | 34  | 35  | 38  | 40  | 40  | 42  | 42  | 39  | 35  | 34  |     |     |     |
| 118 | 120 | 118 | 115 | 113 | 112 | 111 | 112 | 112 | 112 | 110 | 110 | 108 | 104 | 103 | 100 | 105 | 107 | 110 | 117 | 118 | 117 | 117 | 103 | 90  | 83  | 53  | 46  | 39  | 29  | 30  | 31  | 32  | 36  | 37  | 38  | 38  | 38  | 36  | 34  | 33  |     |     |     |
| 110 | 112 | 112 | 111 | 110 | 107 | 106 | 105 | 103 | 103 | 100 | 100 | 97  | 95  | 94  | 88  | 88  | 90  |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |     |

## WHAT ARE PIXELS?

- ▶ the smallest single component of a rasterized digital image
  - ▶ (rasterized meaning matrix-like)
- ▶ the limiting factor in the resolution of an image
- ▶ There are a finite number of pixels in an image
- ▶ Each pixel represents a point sample in the image
  - ▶ Has a discrete coordinate (row, column) in the image
  - ▶ Has a quantized value that indicates brightness

A black and white aerial photograph of a massive concrete dam. The dam is a thick, curved wall with a walkway running along its top edge. Two small figures of people are visible on the walkway, emphasizing the enormous scale of the structure. The surrounding landscape is dark and flat.

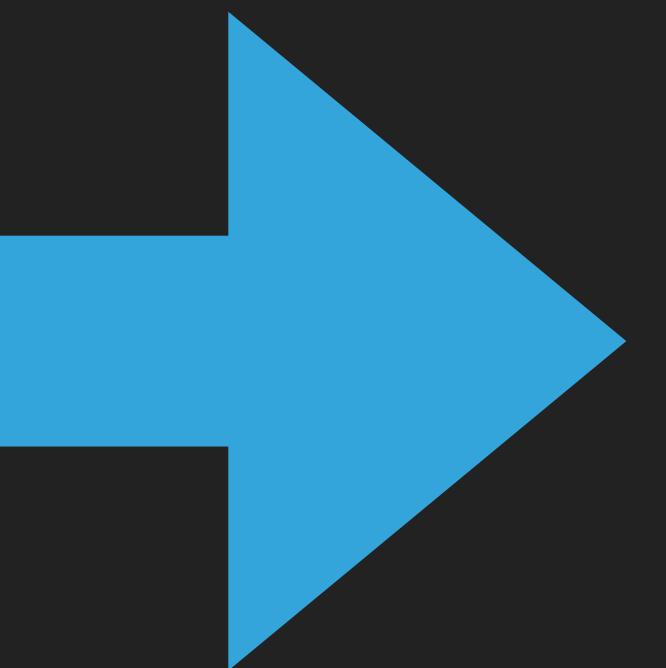
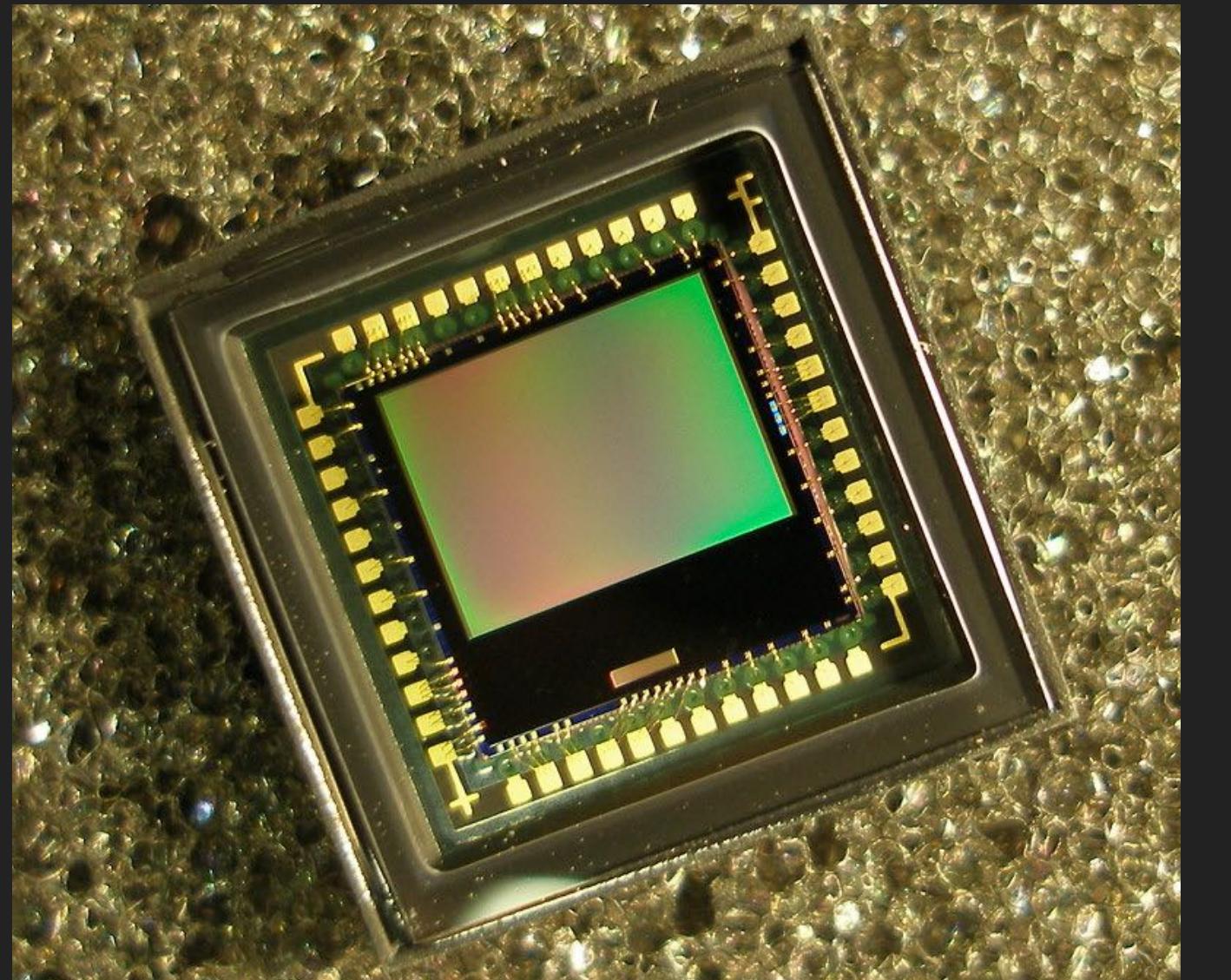
WHY AREN'T THEY CIRCULAR? OR TRIANGULAR?

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WHY ARE IMAGES RECTANGULAR?

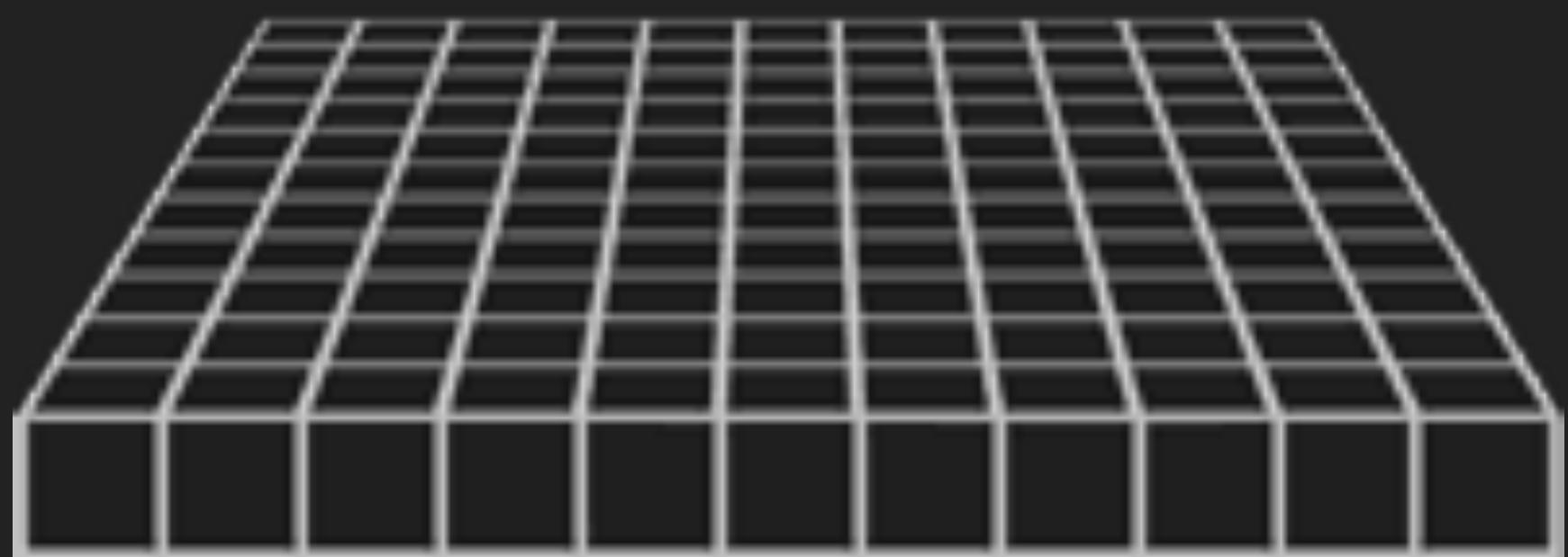
## IMAGE ACQUISITION

# IMAGE SENSORS

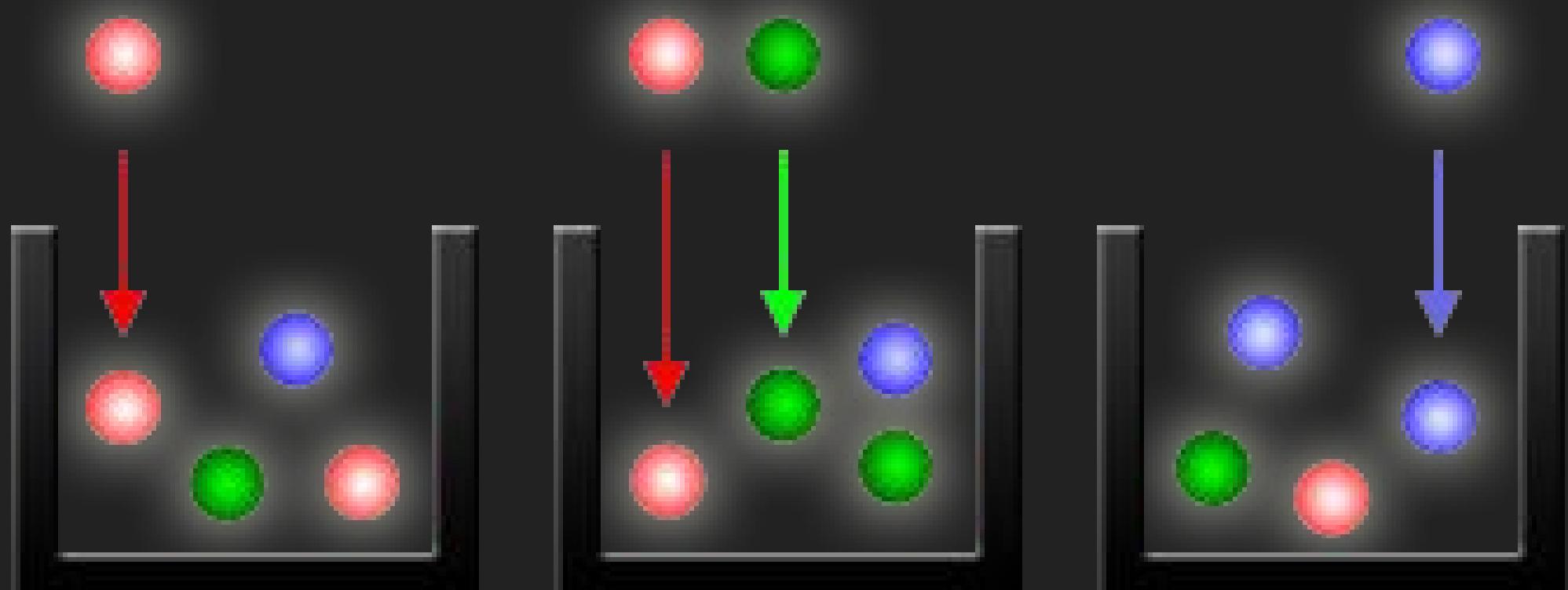


**PHOTOSITES**

aka Pixels

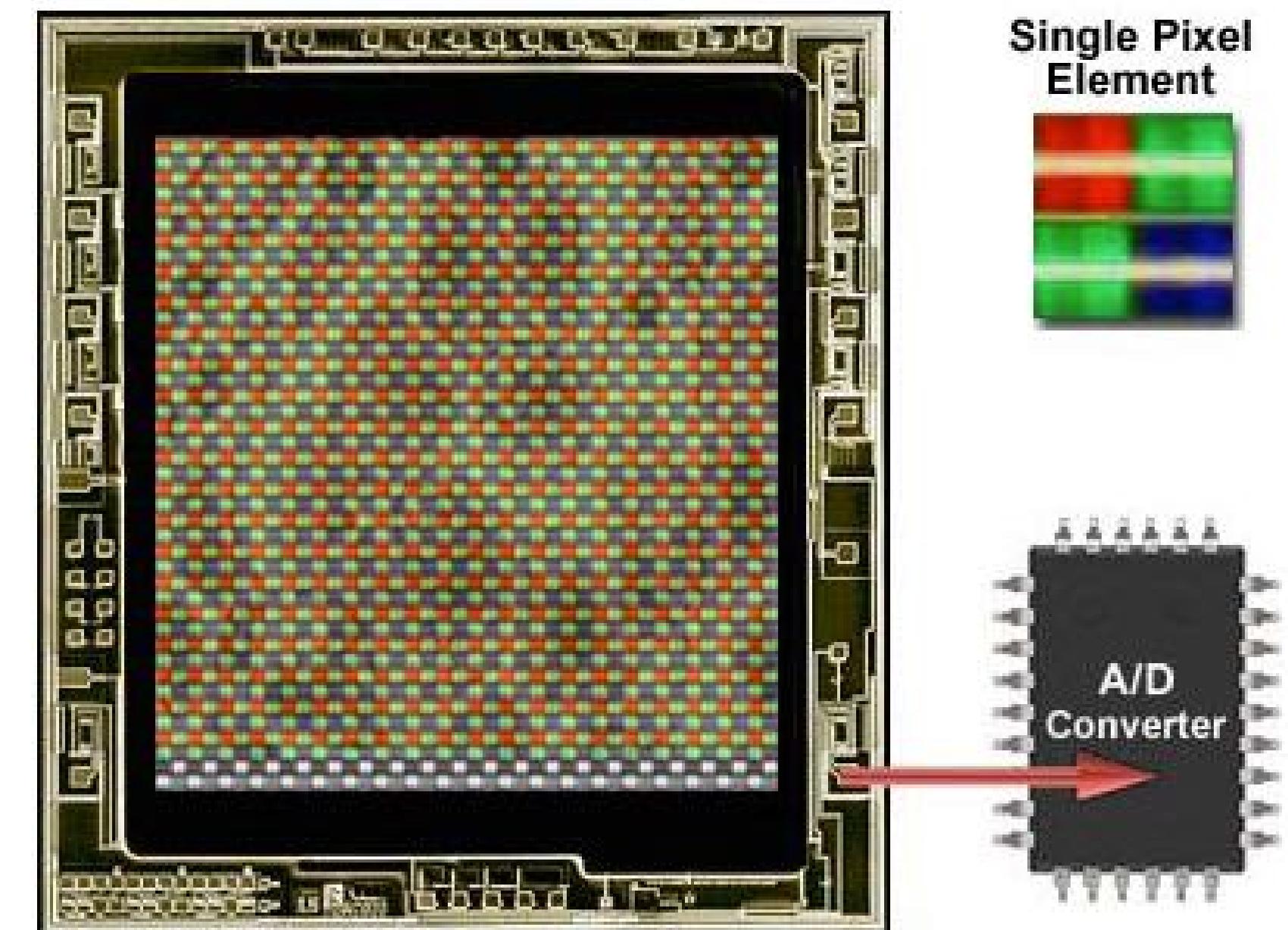


- ▶ Image Sensors made up of a rectangular array of photo sites (aka sensor pixels)
- ▶ Photosites Convert Photons to Electrons (voltage)
- ▶ Number of photo sites = effective resolution of chip



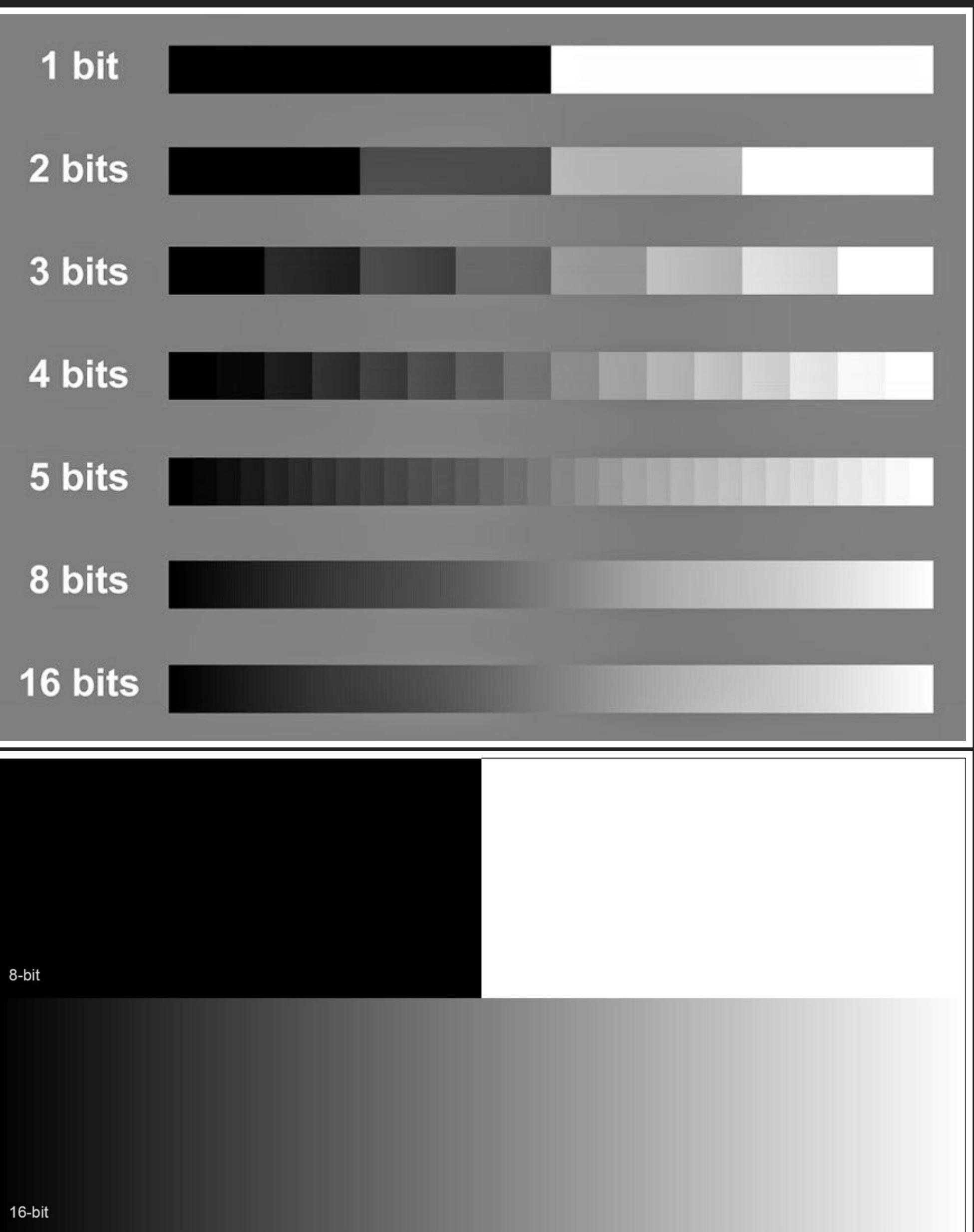
# ANALOG TO DIGITAL CONVERTER

- ▶ Converts continuous photon voltages to quantized integer values
  - ▶ a discrete set of integers (eg 0 - 255) which determines the bit depth of the image
- ▶ More money, more bit-depth.
  - ▶ More expensive sensors are larger with better ADCs which churn out **Higher Bit Depths** with a wide **Dynamic Range**



# WHAT IS BIT DEPTH?

- ▶ Intrinsic property of a raster image that indicates the:
  - ▶ Maximum possible intensity value of a given image
  - ▶ Number of shades of intensity (based on powers of 2)
    - ▶  $8\text{-bit} = 2^8 = 256$  shades of gray
    - ▶  $16\text{-bit} = 2^{16} = 65,536$  shades of gray
- ▶ Not to be confused with Bits and Bytes in Computer Memory
  - ▶  $1 \text{ Byte} = 8 \text{ bits}$  = smallest addressable unit in computer memory
  - ▶ e.g. megabyte, gigabyte



## IMAGE PROPERTIES

### BIT DEPTH



**1-bit**

2 SHADES

**4-bit**

16 SHADES

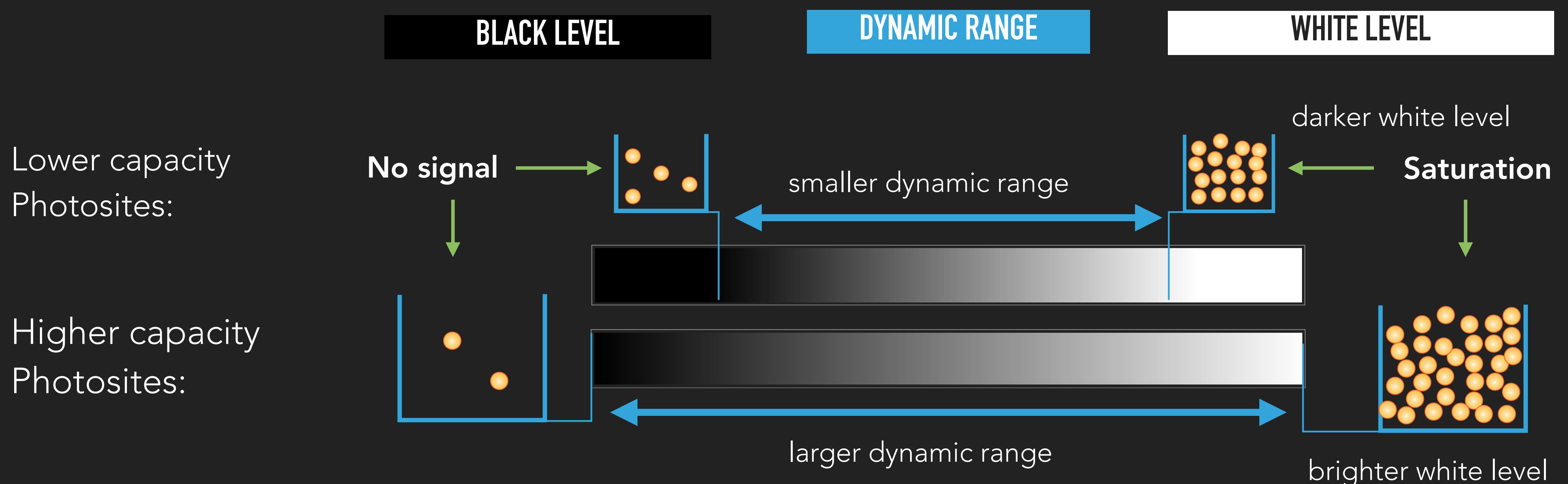
**8-bit**

256 SHADES

- ▶ All images in this example have the same number of pixels, just different bit depth

### WHAT IS DYNAMIC RANGE ?

- ▶ The potential responsive range of a given photosite in an image sensor
- ▶ The range from no signal (black) to saturated signal (white)



## IMAGE ACQUISITION

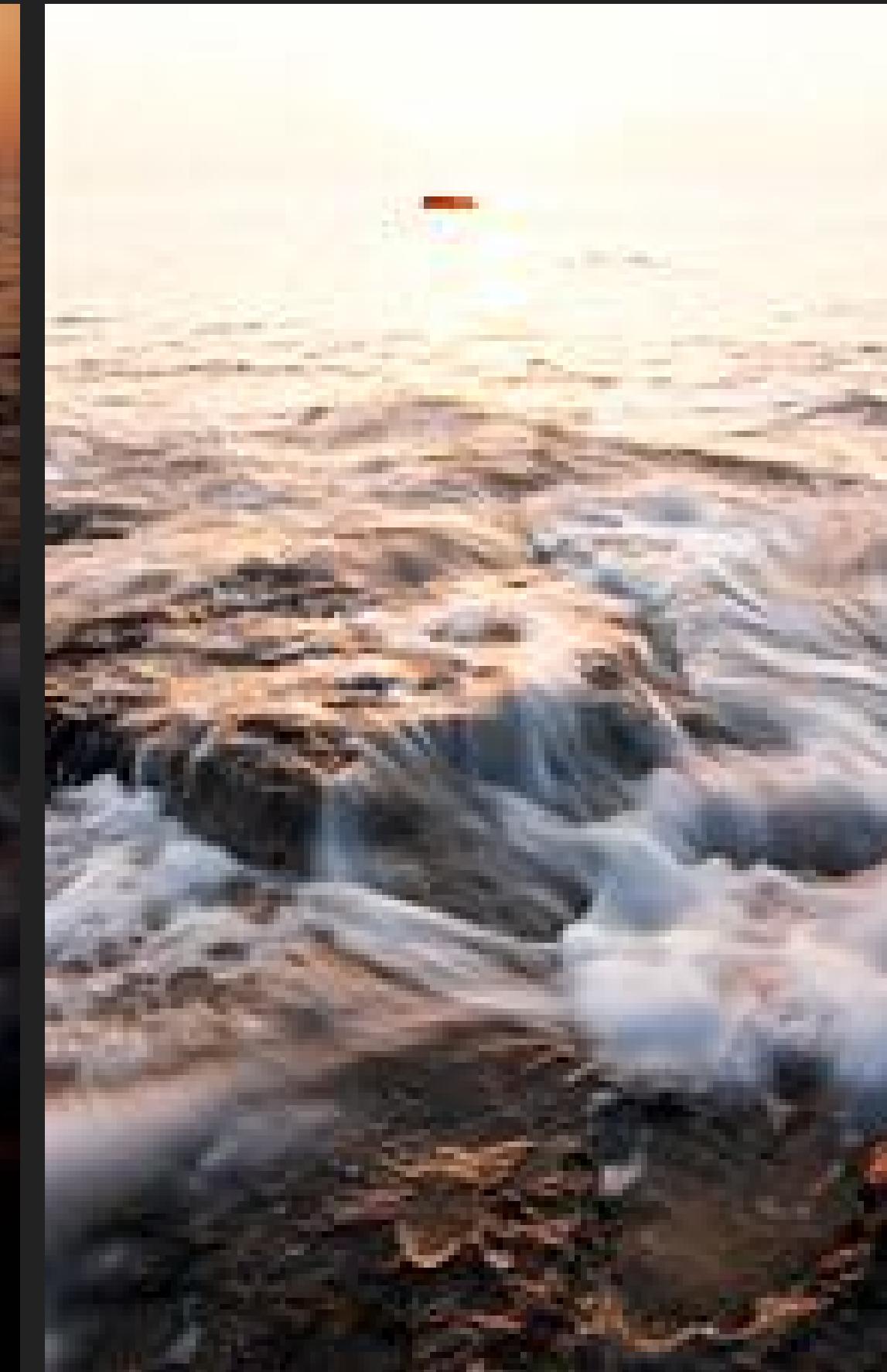
# DYNAMIC RANGE

CAMERA SENSOR



Background in range

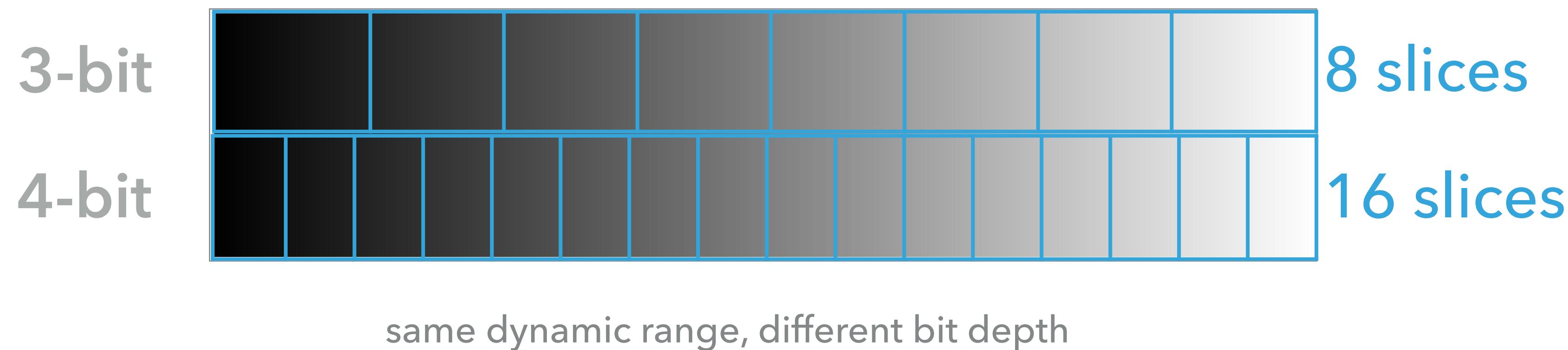
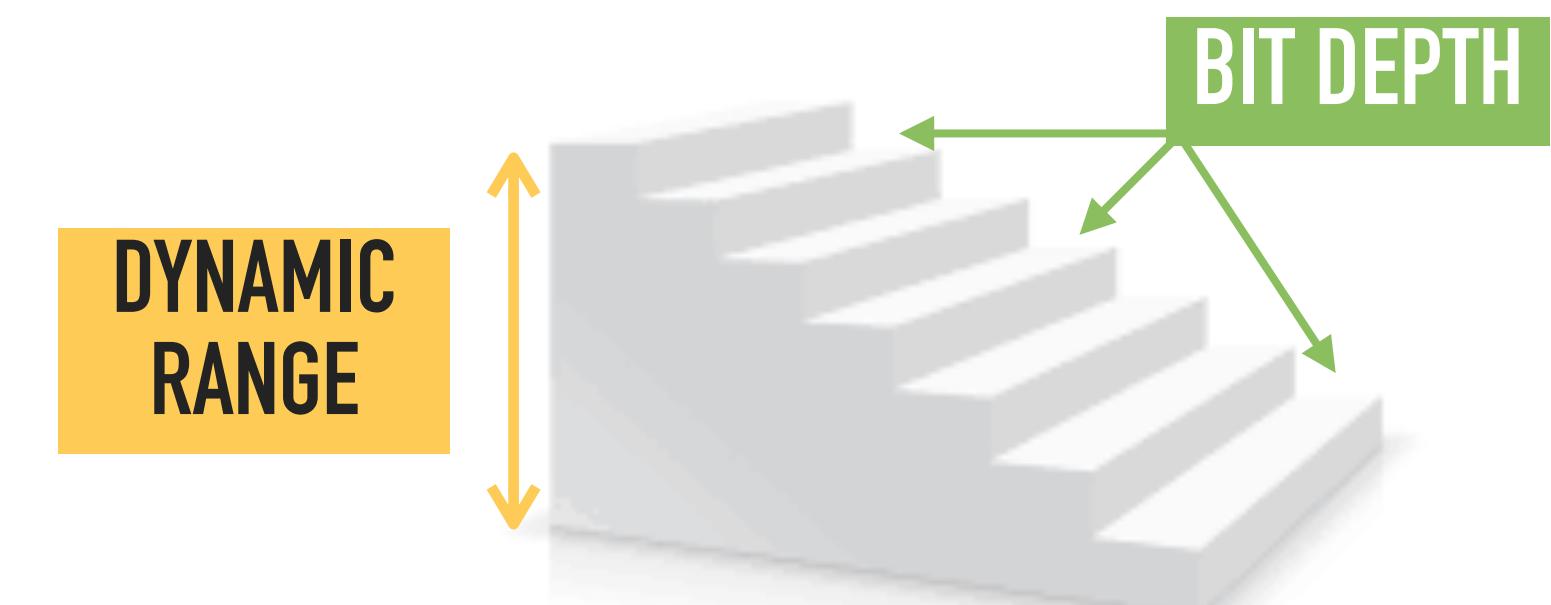
HUMAN EYE

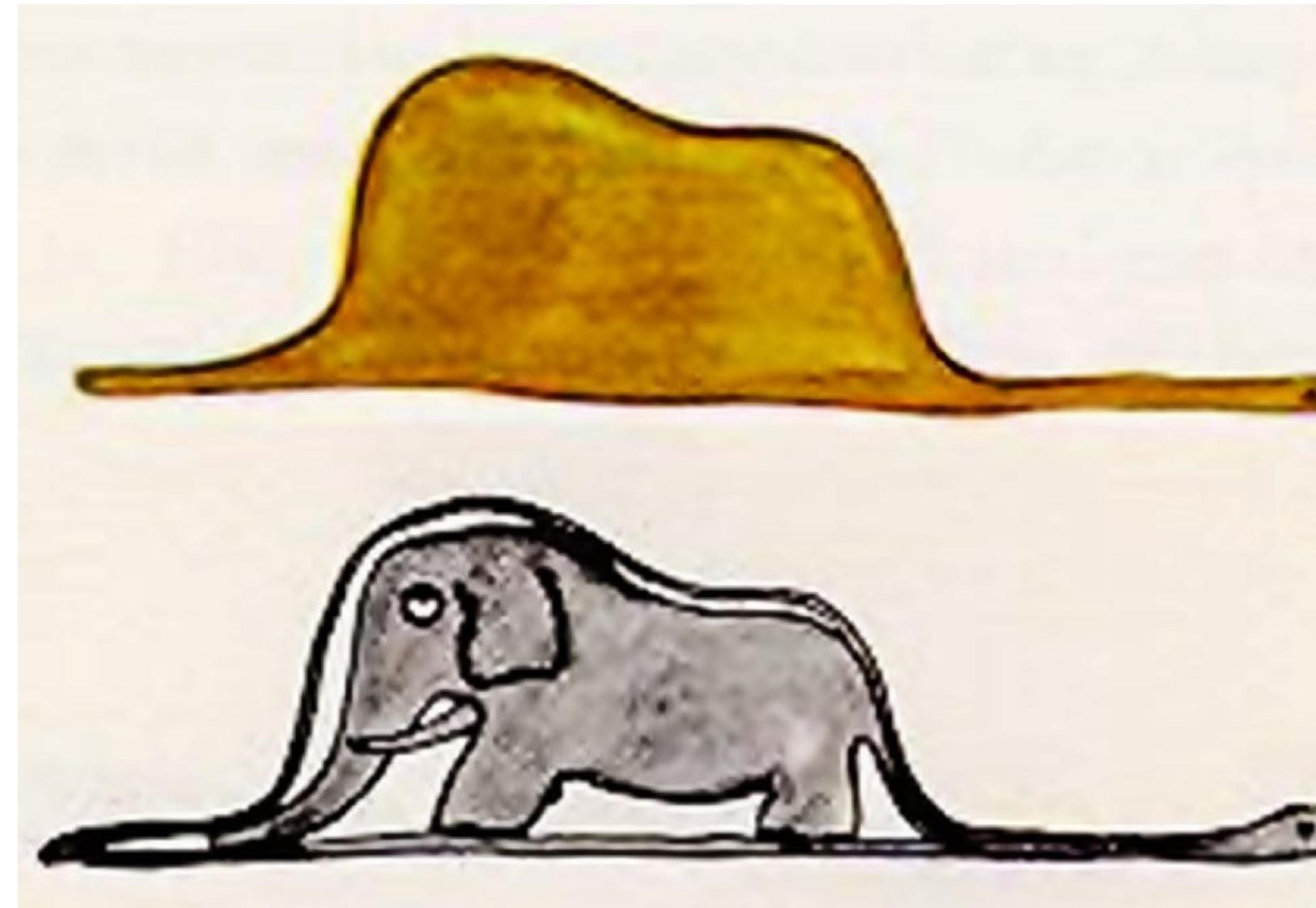


Foreground in range   Greater Dynamic Range

# DYNAMIC RANGE VS BIT DEPTH

- ▶ Dynamic Range is the potential responsive range of a given photosite
- ▶ Bit depth indicates the number of shades into which the dynamic range is chopped (binned):
- ▶ Dynamic range = height of steps, bit depth = number of stairs





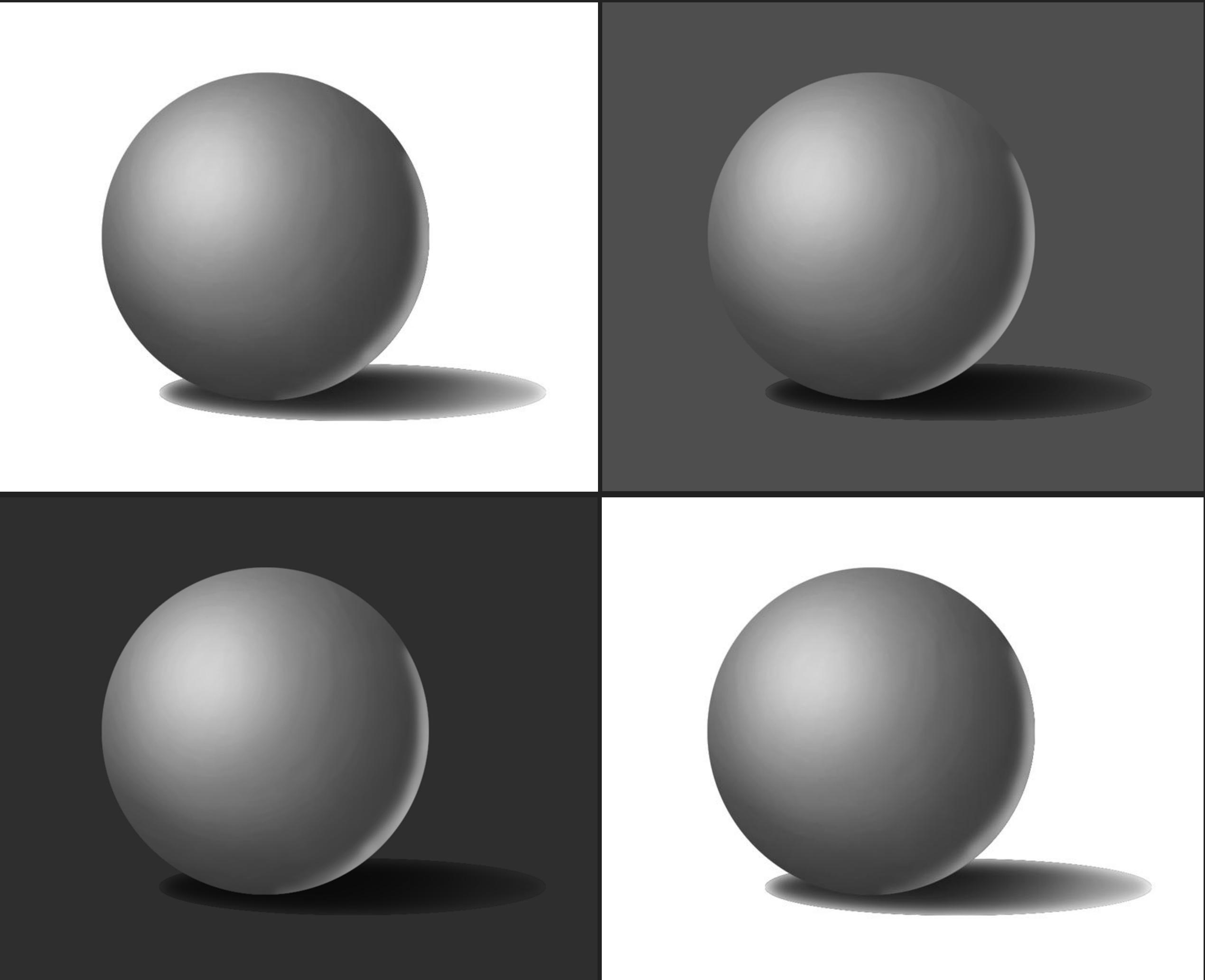
A GOOD IMAGE HAS GOOD CONTRAST

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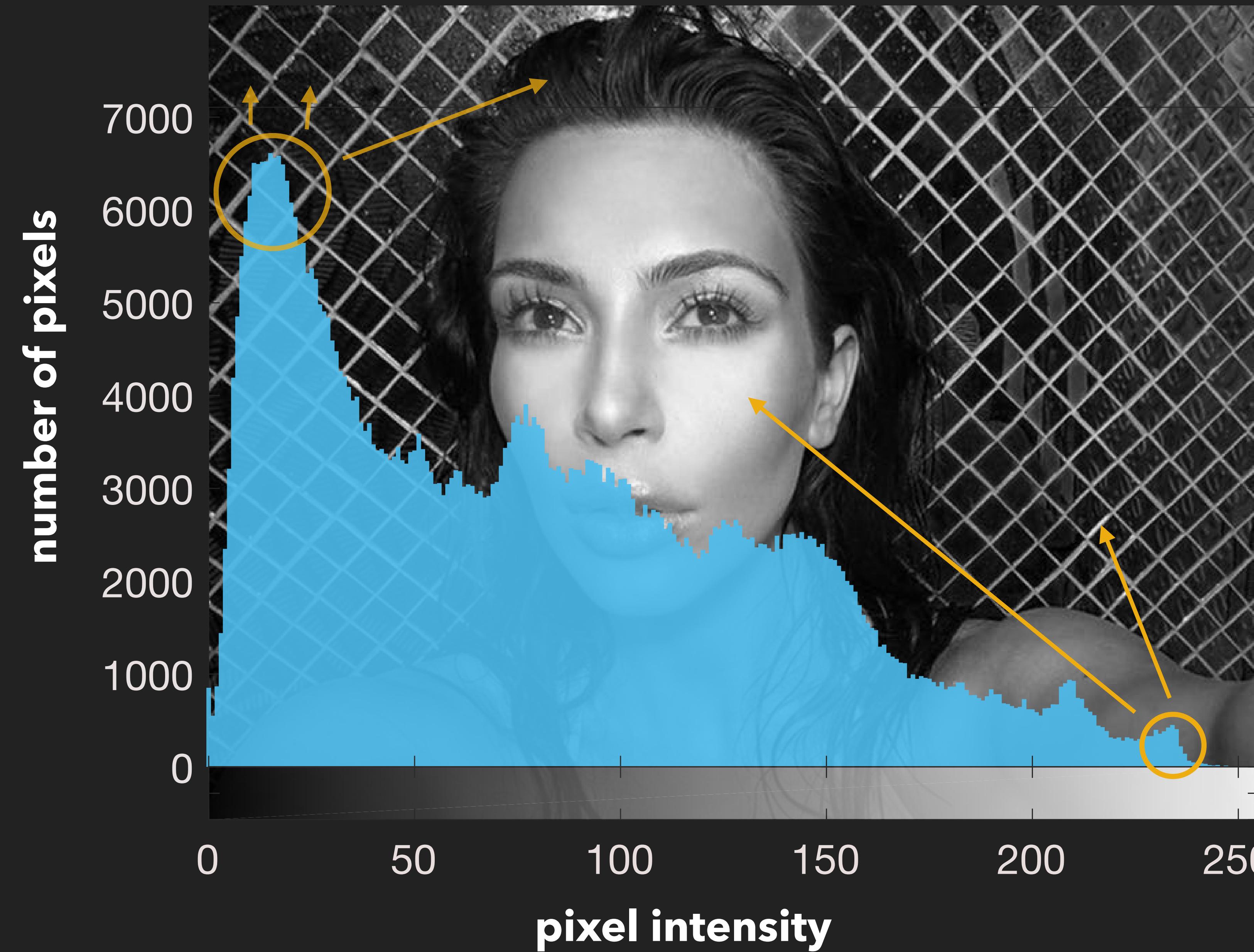
# HISTOGRAMS AND CONTRAST

### CONTRAST

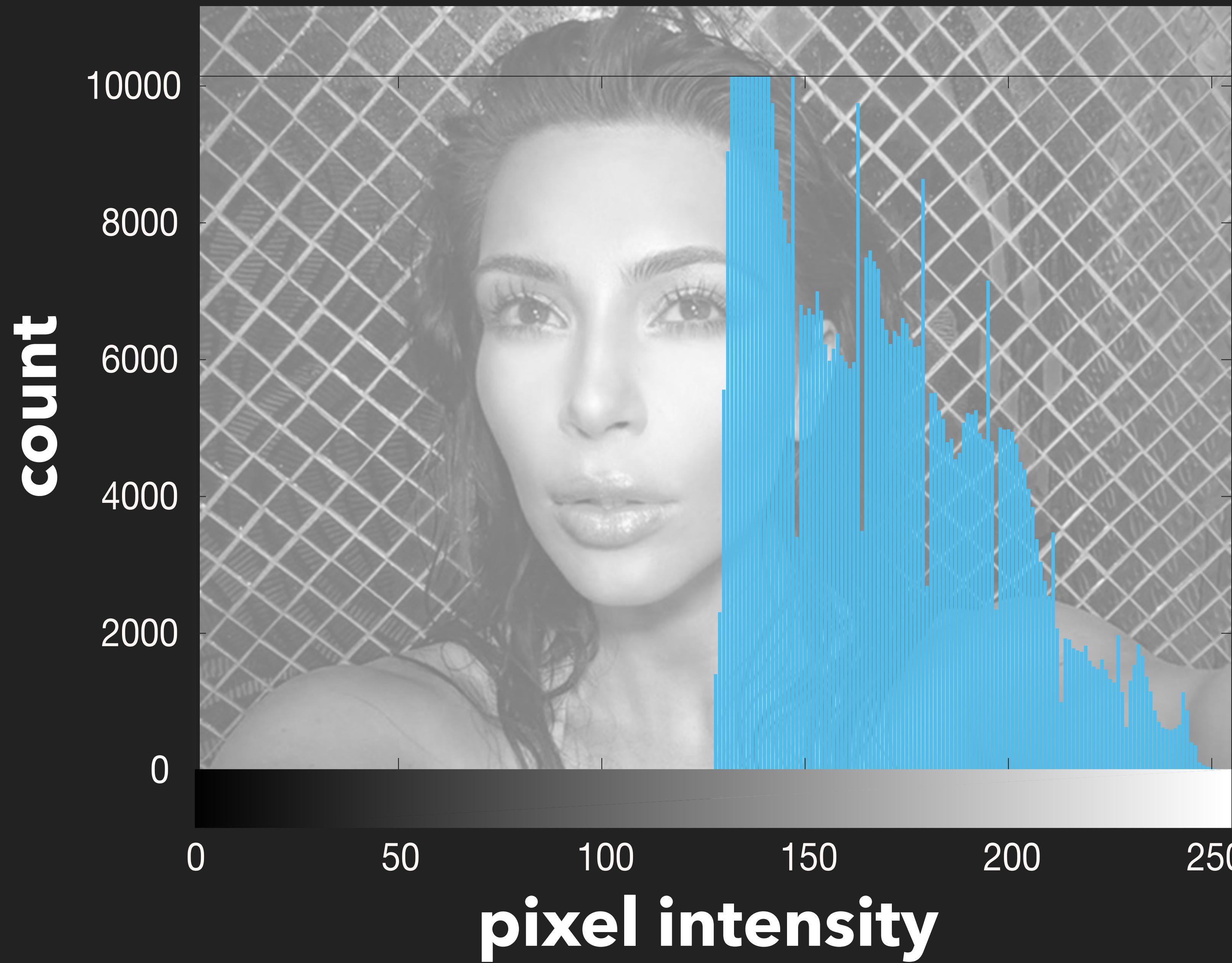
- ▶ Ratio between the brightest and dimmest Pixel
- ▶ Definition a bit imprecise.
- ▶ multiple ways to calculate it
- ▶ Good images have good contrast for analysis



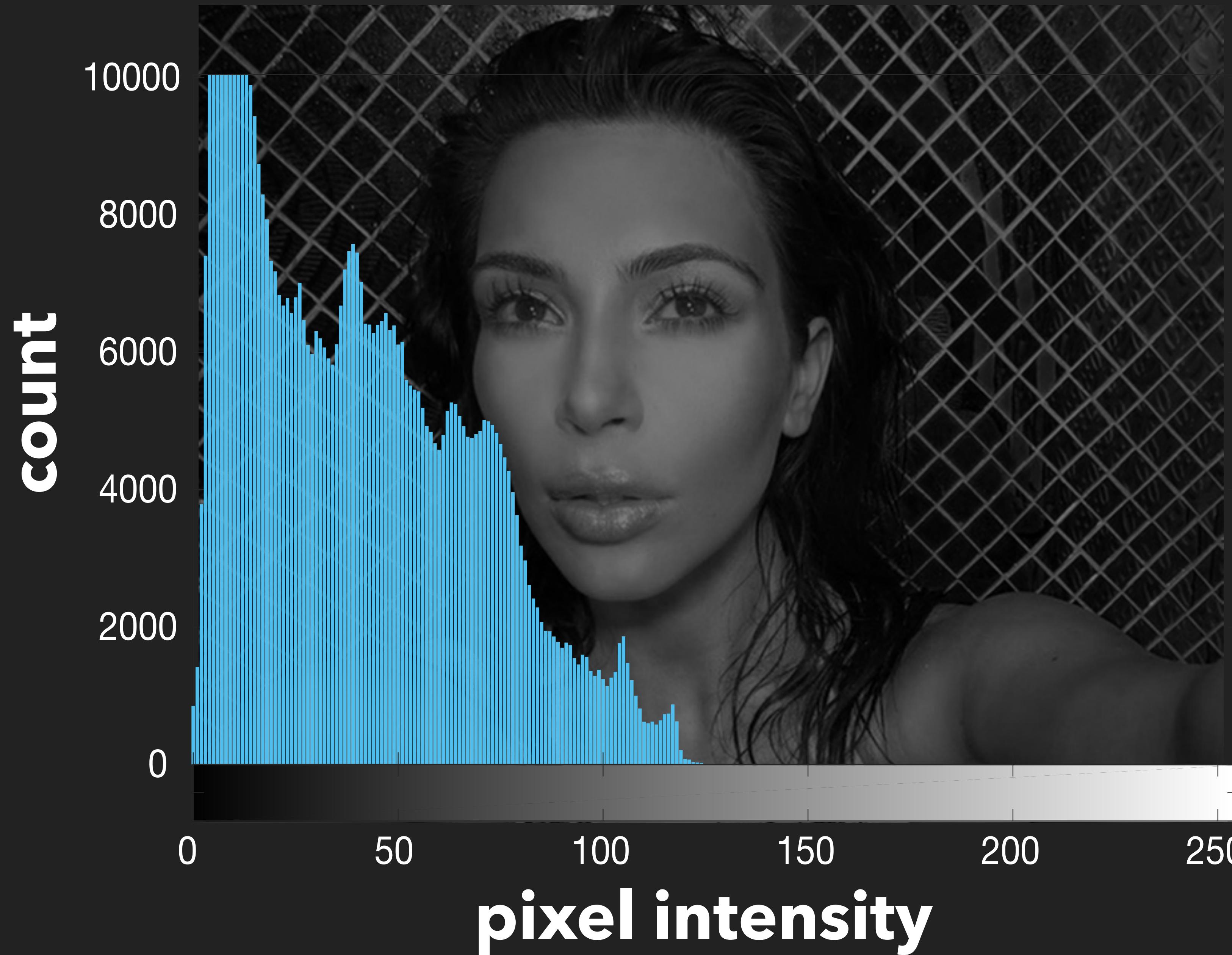
# IMAGE HISTOGRAM - A DISTRIBUTION OF PIXEL INTENSITIES



# POOR CONTRAST: TOO MANY BRIGHT AND NO DARK PIXELS



# POOR CONTRAST: TOO MANY DARK AND NO BRIGHT PIXELS

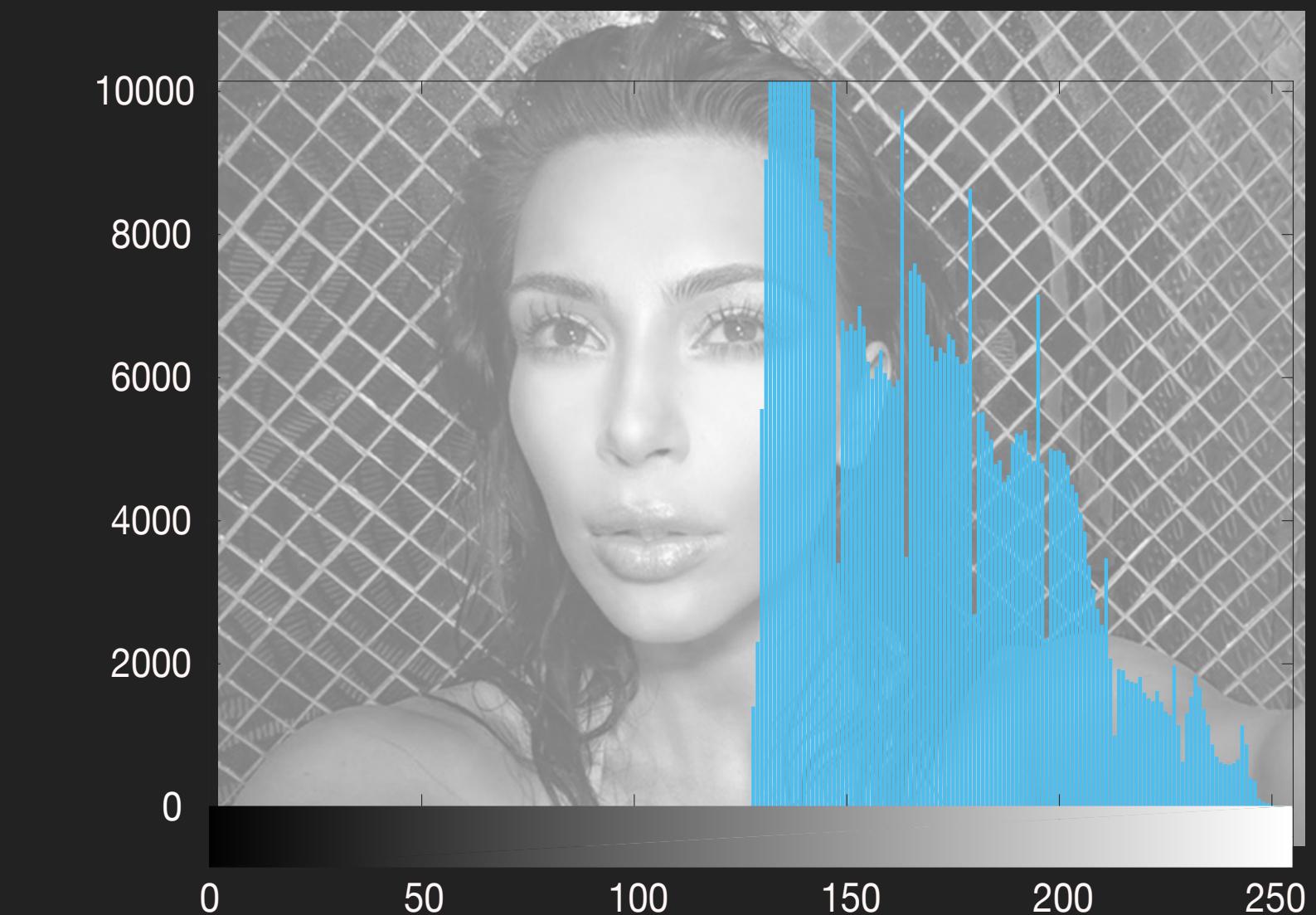


## HISTOGRAMS AND CONTRAST

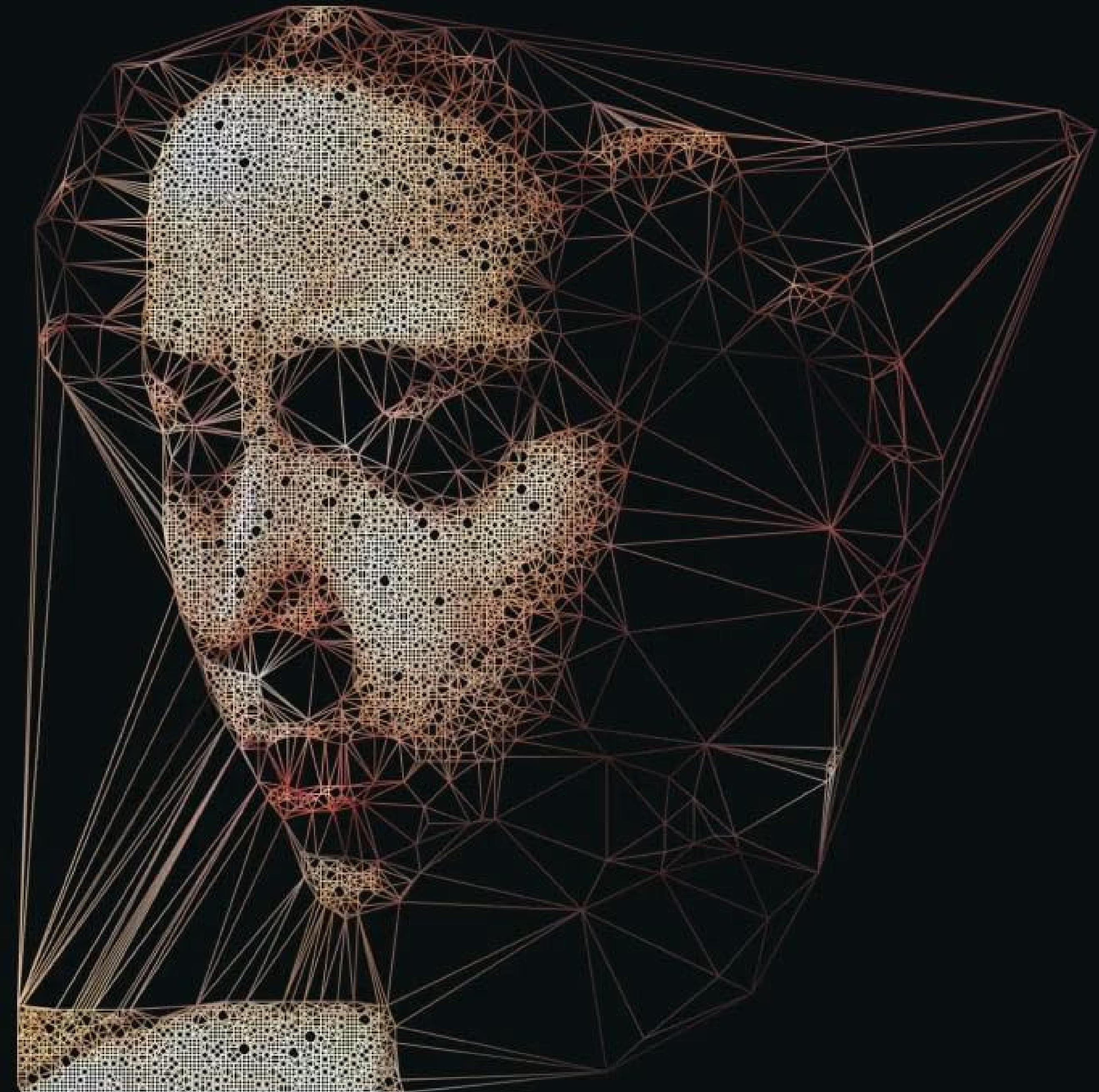
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### TAKE-AWAY

- ▶ Make sure your images have a nice distribution of pixel intensity across the entire dynamic range of the image before you process the image



# IMAGE PROCESSING



# WHAT IS IMAGE PROCESSING?

- ▶ Ultimately boils down to manipulate three properties:

## 1. Pixel Values

- ▶ e.g. contrast enhancement, filtering, thresholding

## 2. Pixel Location

- ▶ e.g. ROIs, cropping, segmentation

## 3. Pixel Size

- ▶ e.g. calculating area based on pixel dimensions, removing objects that are too big/too small

## COMMON STEPS

- ▶ Enhance Contrast
- ▶ Filter
- ▶ Threshold
- ▶ Morphological Operations
- ▶ Region Properties
- ▶ Data analysis

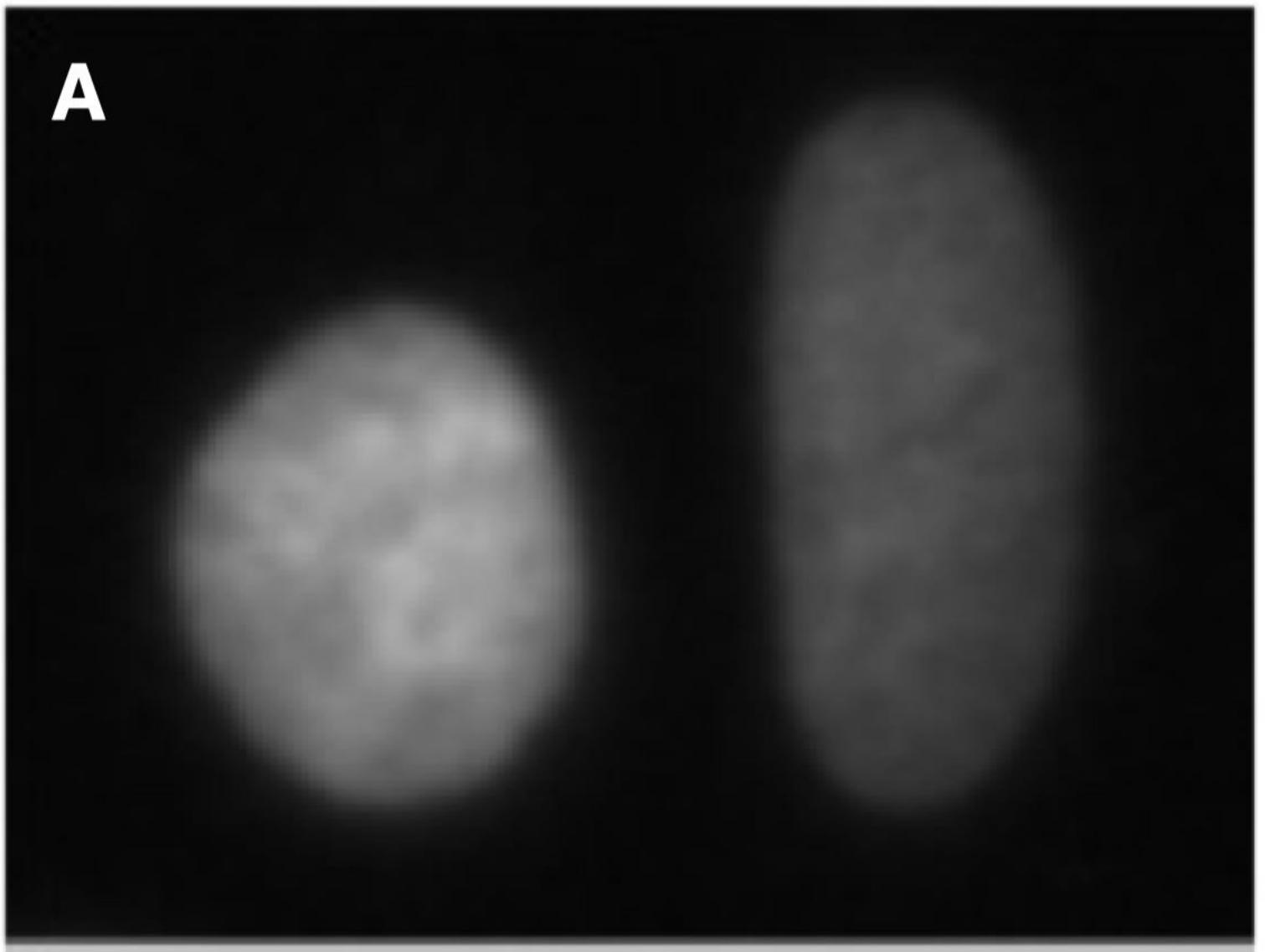


# SCIENTIFIC MALFEASANCE

# BAD PHOTOSHOP

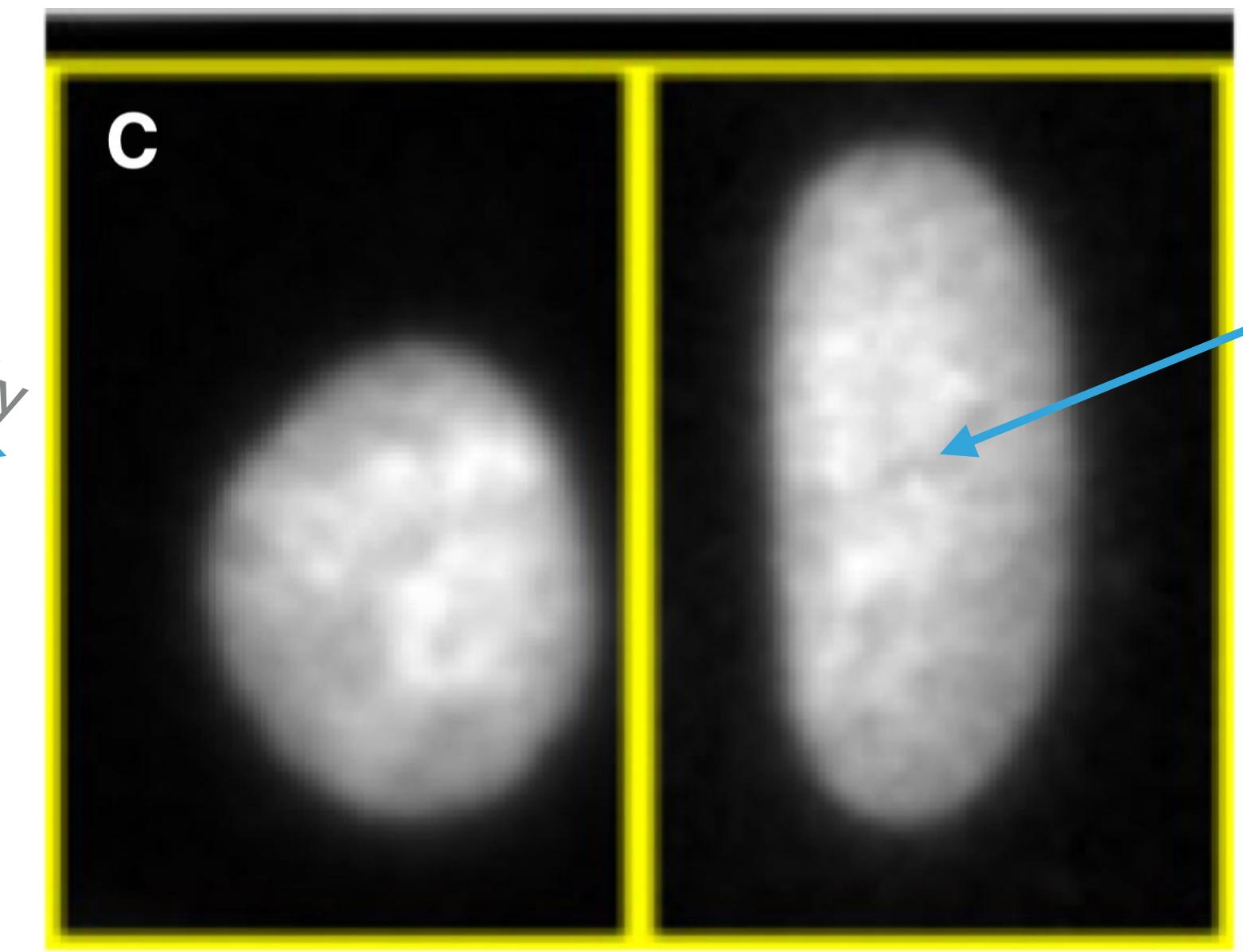
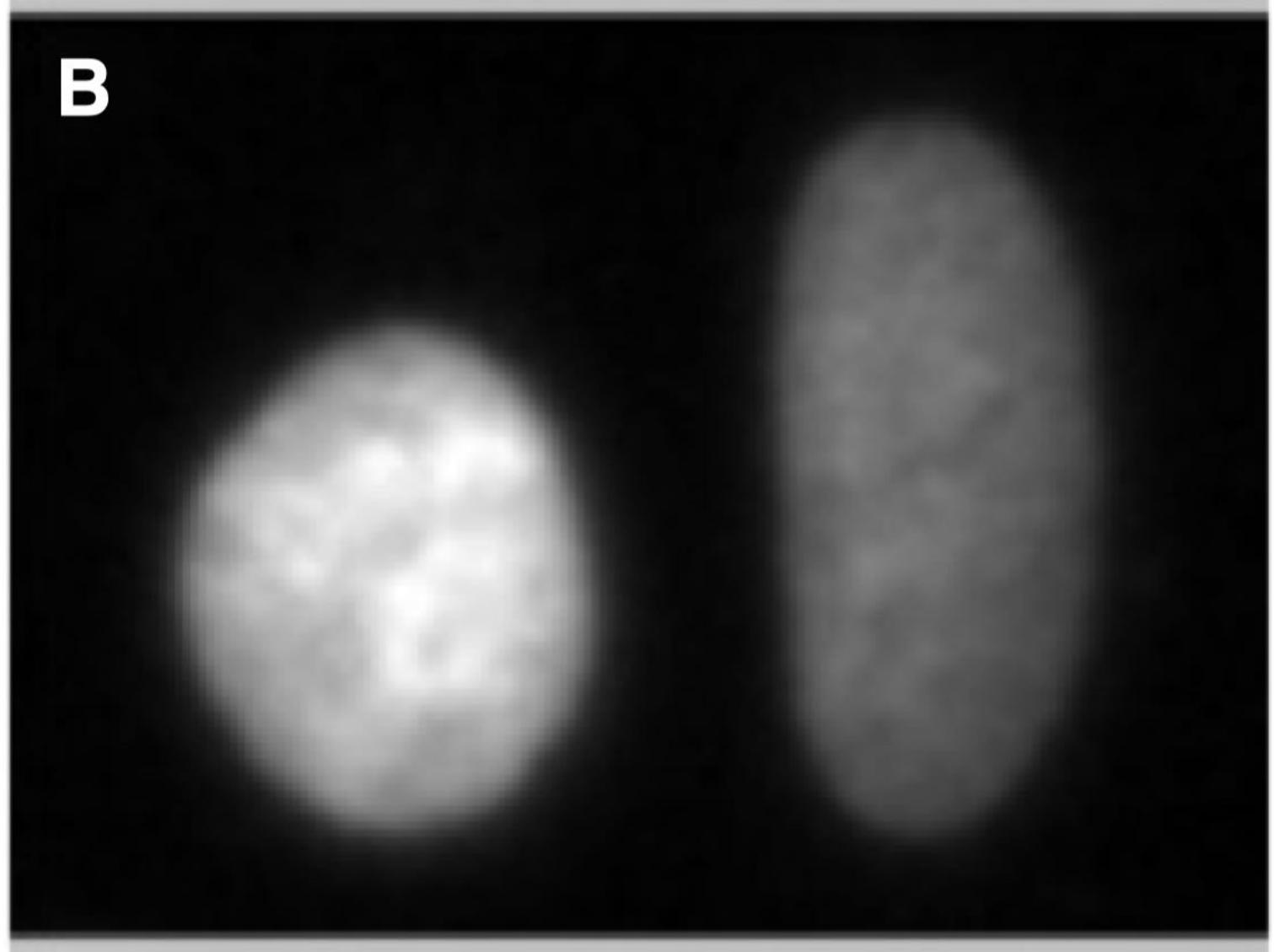


## THE WRONG WAY TO ENHANCE CONTRAST



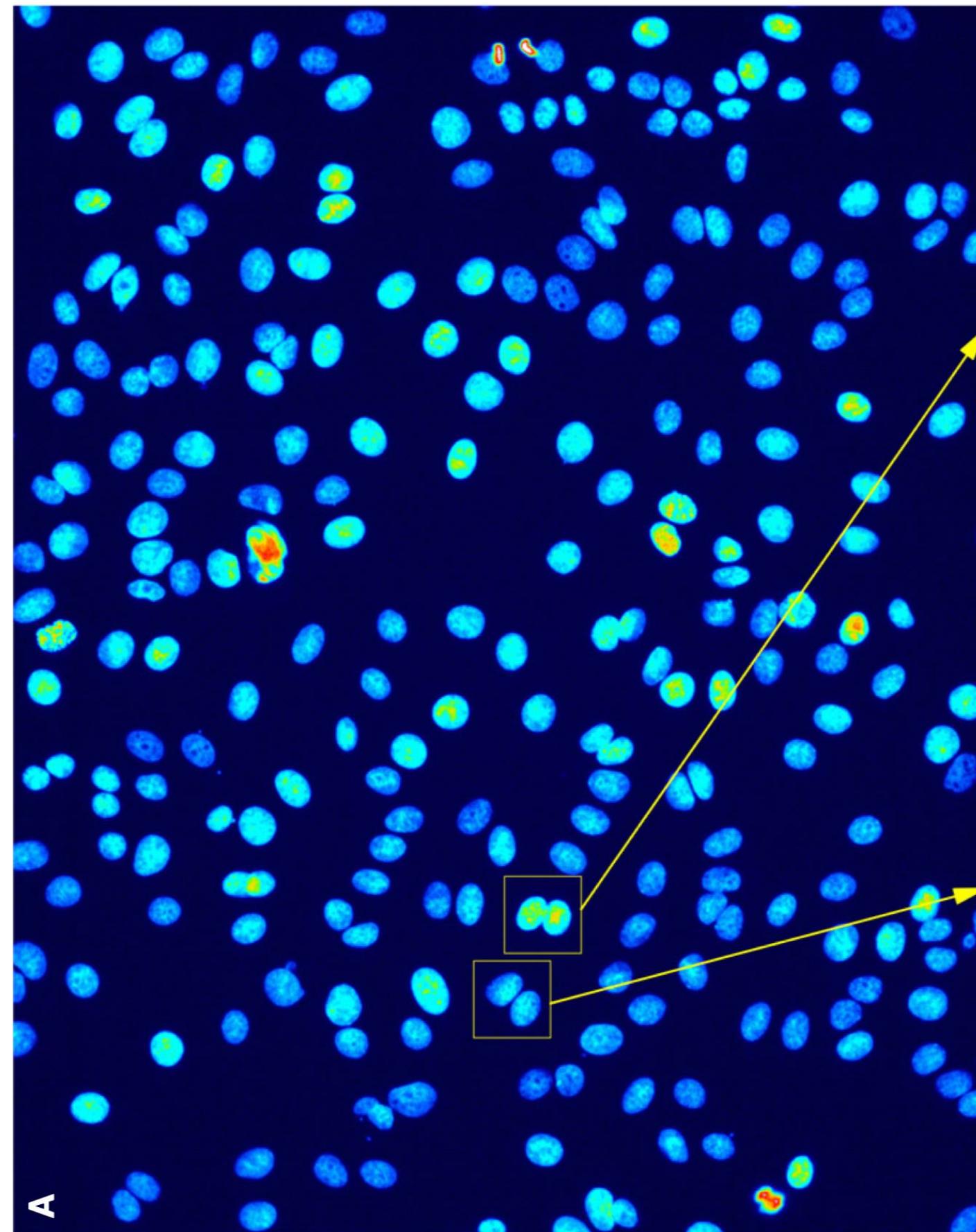
Good - intensity adjusted together

Bad - intensity adjusted separately

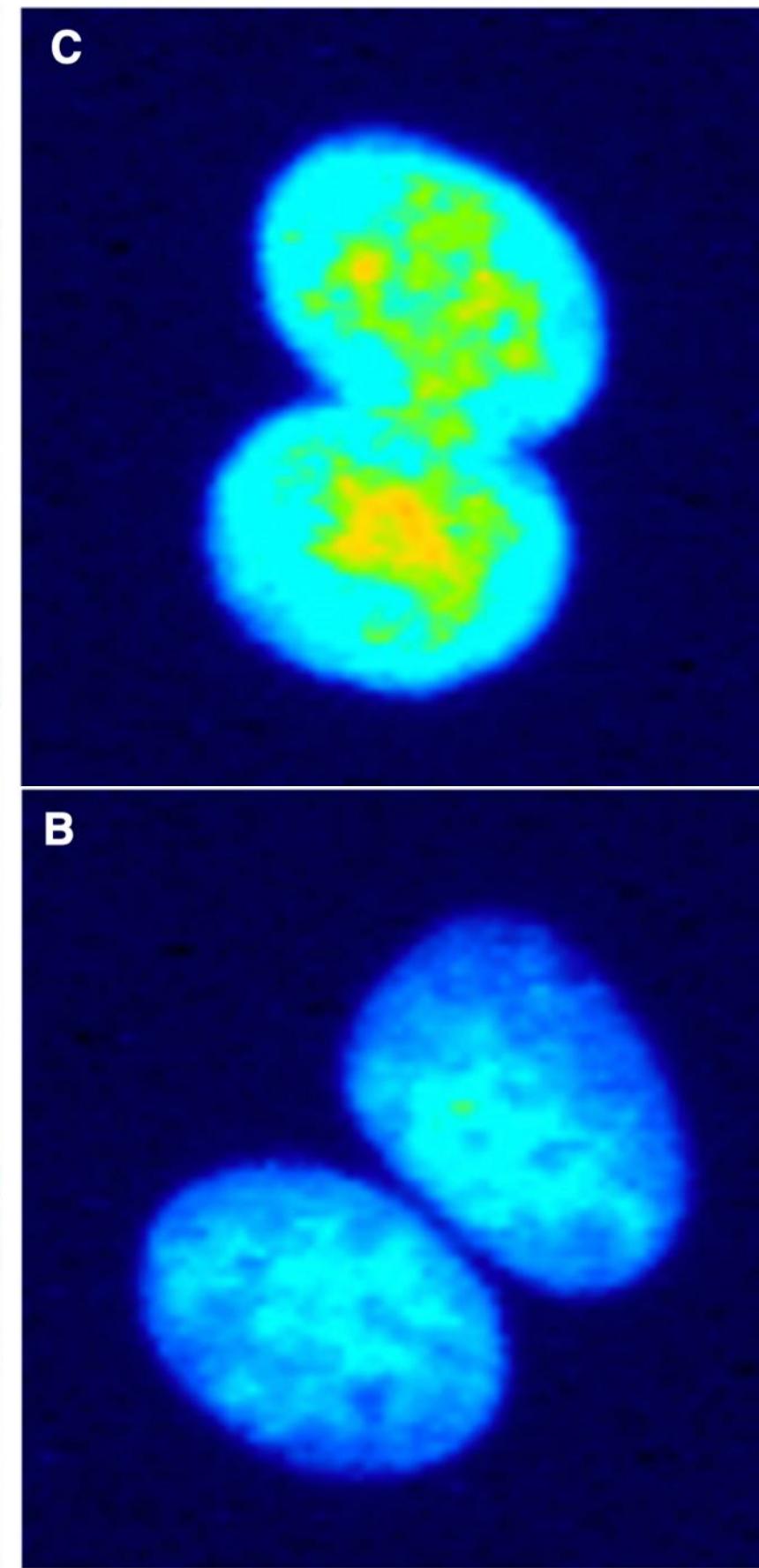


# WRONG WAY TO CONVERT BIT-DEPTHS AND NORMALIZE

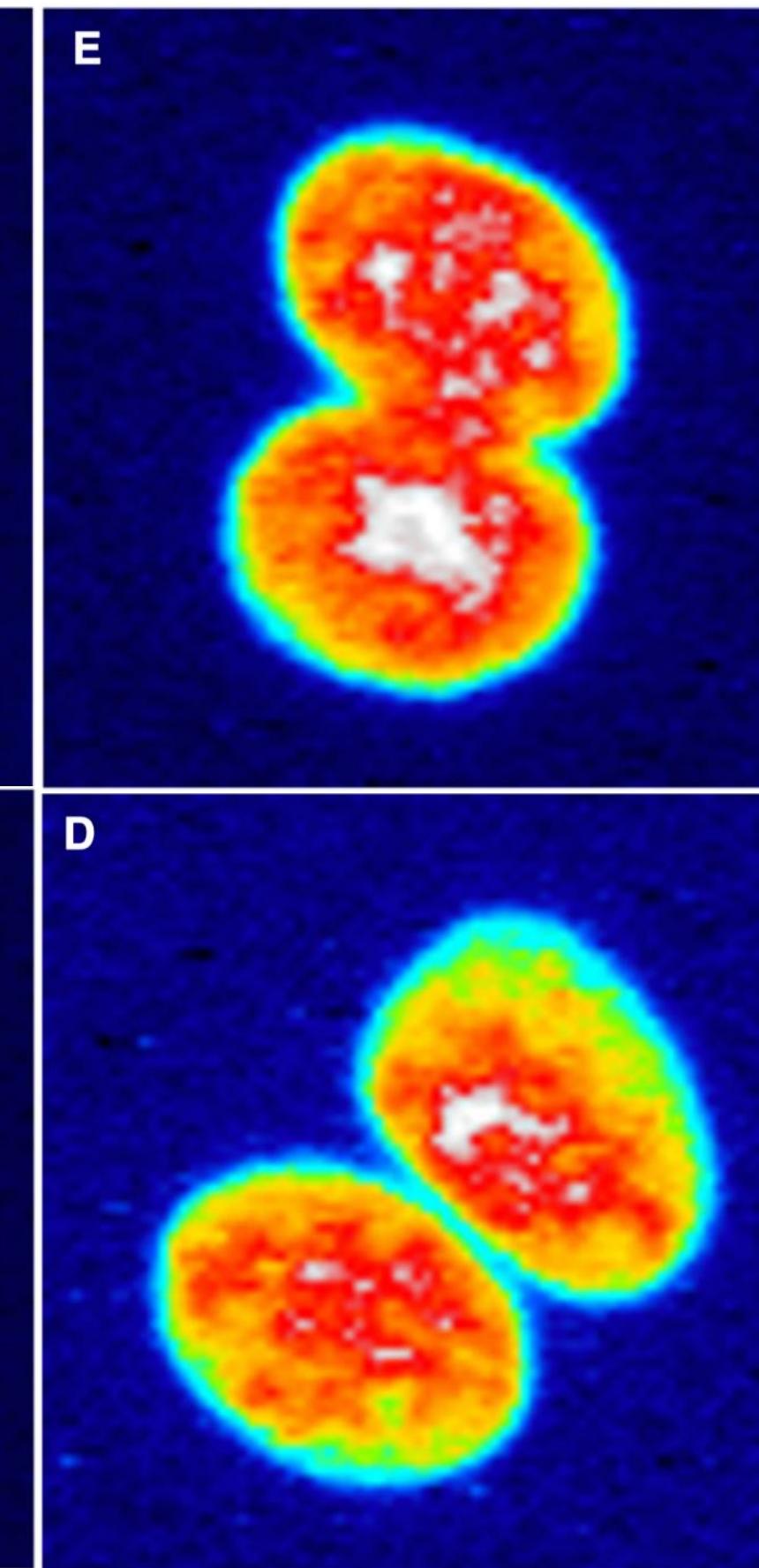
Original Image



Correct Way



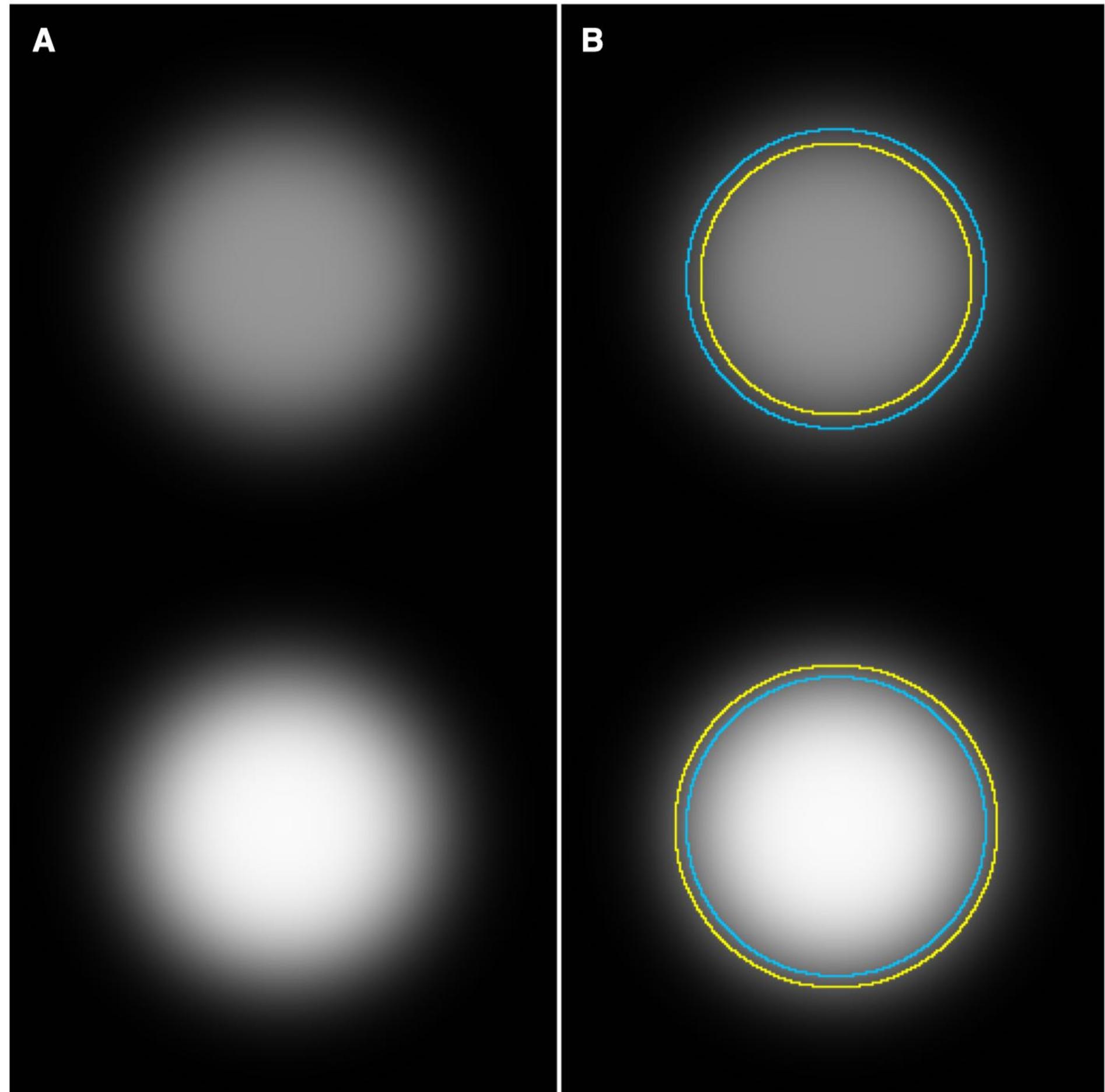
Incorrect Way



- ▶ **Correct way:** C and B were first normalized together and then cropped
  - ▶ still have evident intensity differences
- ▶ **Wrong Way:** D and E were first cropped and then normalized independently
  - ▶ they look now look equally bright

### WRONG WAY TO SEGMENT

- ▶ Automated segmentation tool used to capture dots captured different sized ROIs (regions-of-interests)
- ▶ **Note** Blue is outside Yellow in top image and vice versa in lower image
- ▶ Same colored Circles have the same mean intensity value across the dots
- ▶ So, depending on how you segment the dots, you can get varying results



# BEST PRACTICES

## IMAGE PROCESSING PROTOCOLS

- ▶ Is there a basic protocol all image processing can follow?
  - ▶ Not really
  - ▶ Image Processing is diverse and specific to the research question
- ▶ Should you follow the same image processing protocol for a given Experiment?
  - ▶ Yes.
  - ▶ For a given experiment, anything you do to the control image, you should do to the experiment image
    - ▶ if you do this ONE simple thing, then you really shouldn't have an issue

## BEST PRACTICES

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# SIMPLE IMAGE PROCESSING CHECKLIST

- ▶ Review the histogram on Acquisition
  - ▶ are the pixels intensities evenly distributed?
  - ▶ are there any saturated pixels
  - ▶ is your signal well above the noise?
  - ▶ does the bit-depth of the image match the bit depth of the acquisition?
- ▶ File Formats - use proprietary or tifs
- ▶ Processing
  - ▶ Correct Bit-depth in the same fashion across all images
  - ▶ adjust the histogram in the same fashion across all images
  - ▶ apply the same filters to ALL the data in the same fashion
    - ▶ eg. do not enhance dim experimental images without enhance control images in the same fashion
- ▶ Ensure your Segmentations are comparing the same things – small differences in mask sizes or locations can result in large differences in results

## TRANSPARENCY: REPORT ALL OF YOUR PROCESSING STEPS

- ▶ Outline the image processing workflow
- ▶ Detail the software used
  - ▶ Open-source preferable to Commercial Software (but use the best software for the job)
  - ▶ Scriptable (Fiji, MATLAB) preferred over simply clicking on menus and entering values (eg Photoshop ,Powerpoint)
- ▶ Create and provide a detailed protocol or script for steps need to recreate the processing
- ▶ List a table of parameters
- ▶ details of the image acquisition protocols: space-time pixel resolution, exposure parameters, microscope settings, etc
- ▶ Upload data and scripts to on-line site, like git-hub

IMAGE PROCESSING

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COLOR

## COLOR MODE

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# COLOR MODE

Bitmap



Grayscale



RGB



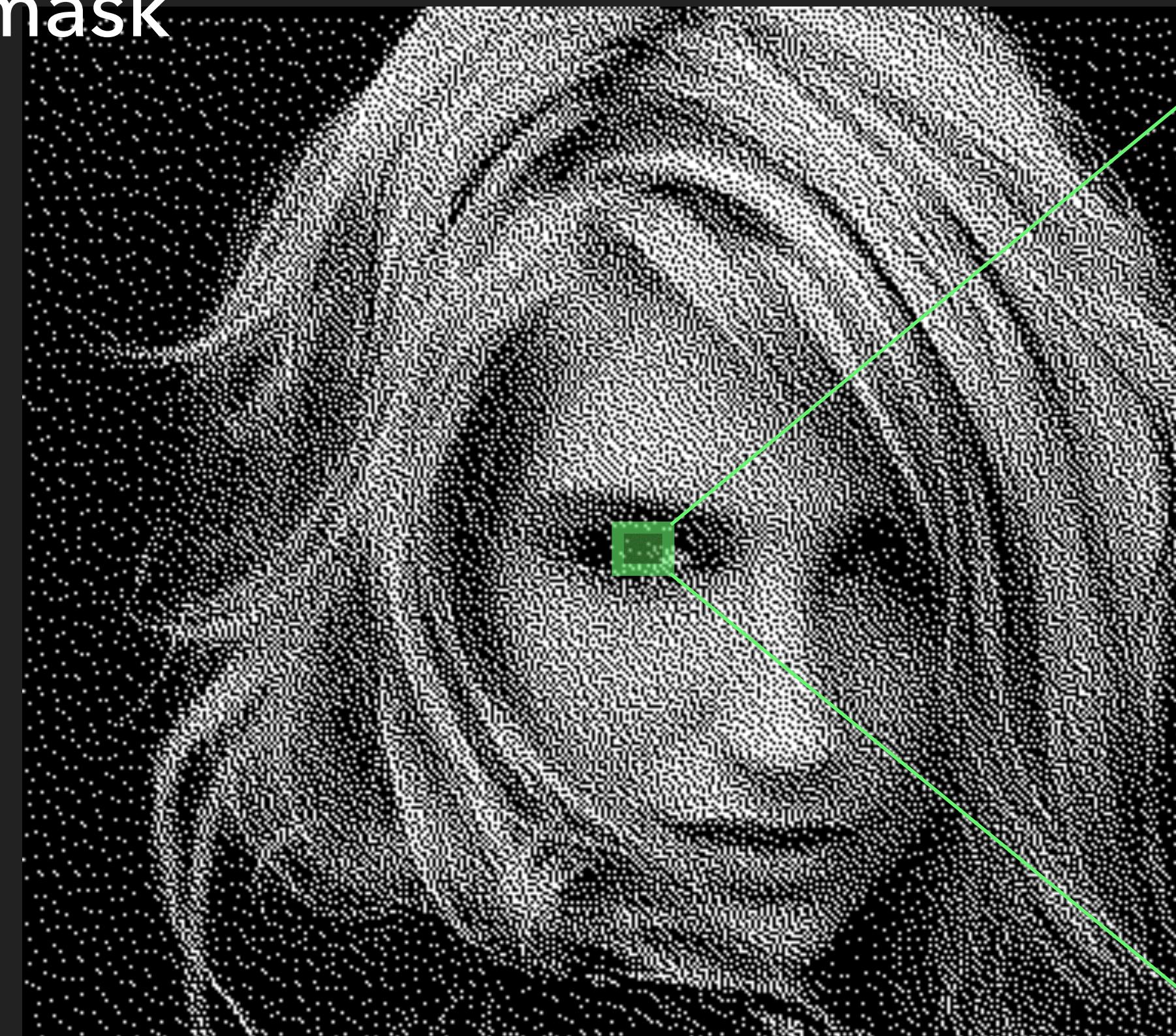
- ▶ Color mode determines how the components of a color are combined, based on the number of color channels in the color model.

## COLOR MODE: BLACK AND WHITE

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# BINARY IMAGE

► a.k.a mask



Pixel Region (Image Tool 1)

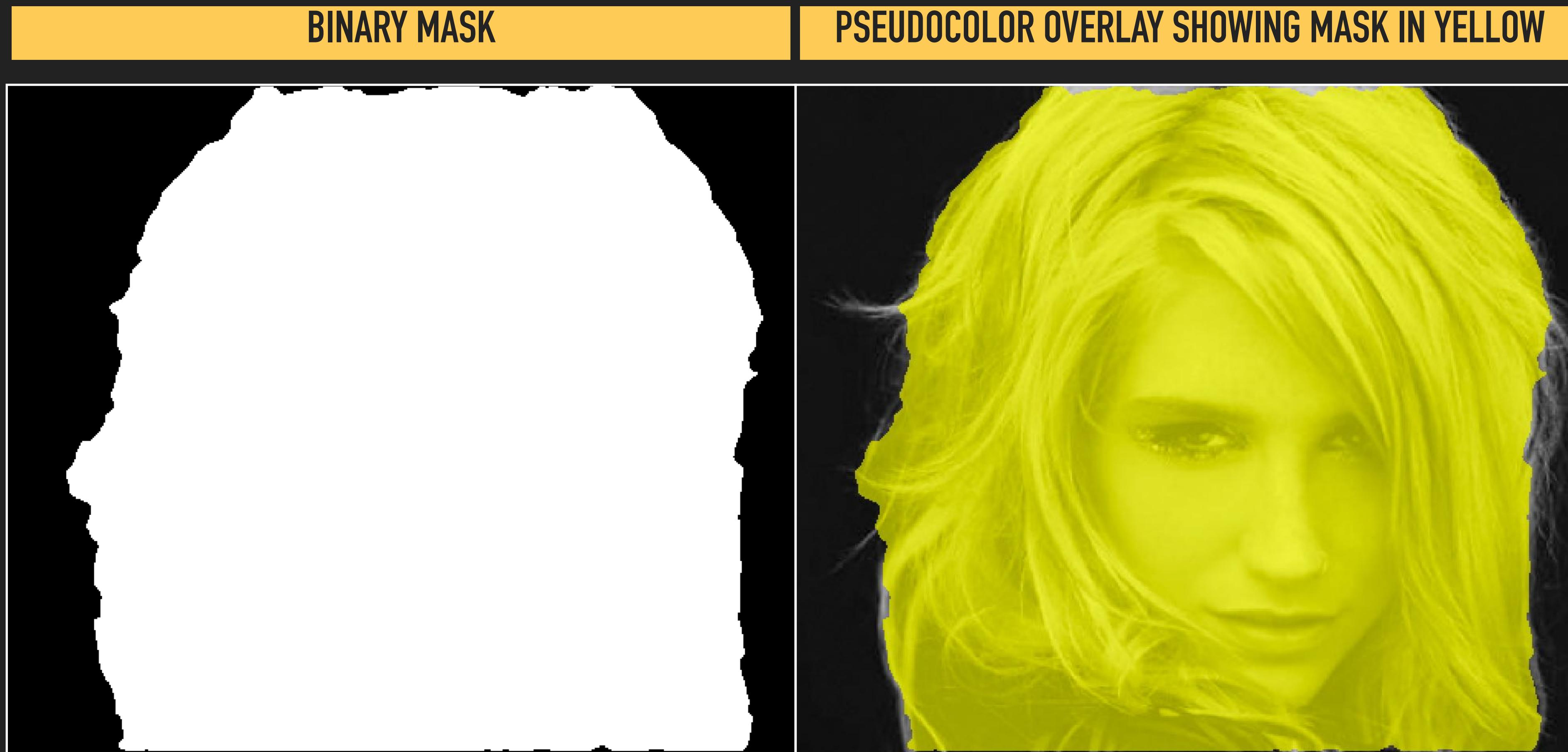
File Edit Window Help

Pixel Info: (230, 172) 1

| 0 | 0 | 1 | 0 | 0 | 1 | 0 | 1 | 1 | 0 | 0 | 1 | 0 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 0 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 0 |
| 0 | 1 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 1 | 0 | 1 |
| 0 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 1 | 0 | 0 | 1 | 0 |
| 0 | 1 | 0 | 1 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 1 | 0 |
| 0 | 0 | 0 | 0 | 1 | 0 | 1 | 0 | 0 | 0 | 0 | 1 | 0 |
| 0 | 1 | 0 | 0 | 0 | 0 | 0 | 0 | 1 | 0 | 1 | 0 | 1 |
| 0 | 0 | 0 | 0 | 1 | 0 | 0 | 1 | 0 | 0 | 0 | 0 | 0 |
| 0 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 1 | 0 | 1 | 0 |
| 0 | 1 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 1 | 0 | 0 |

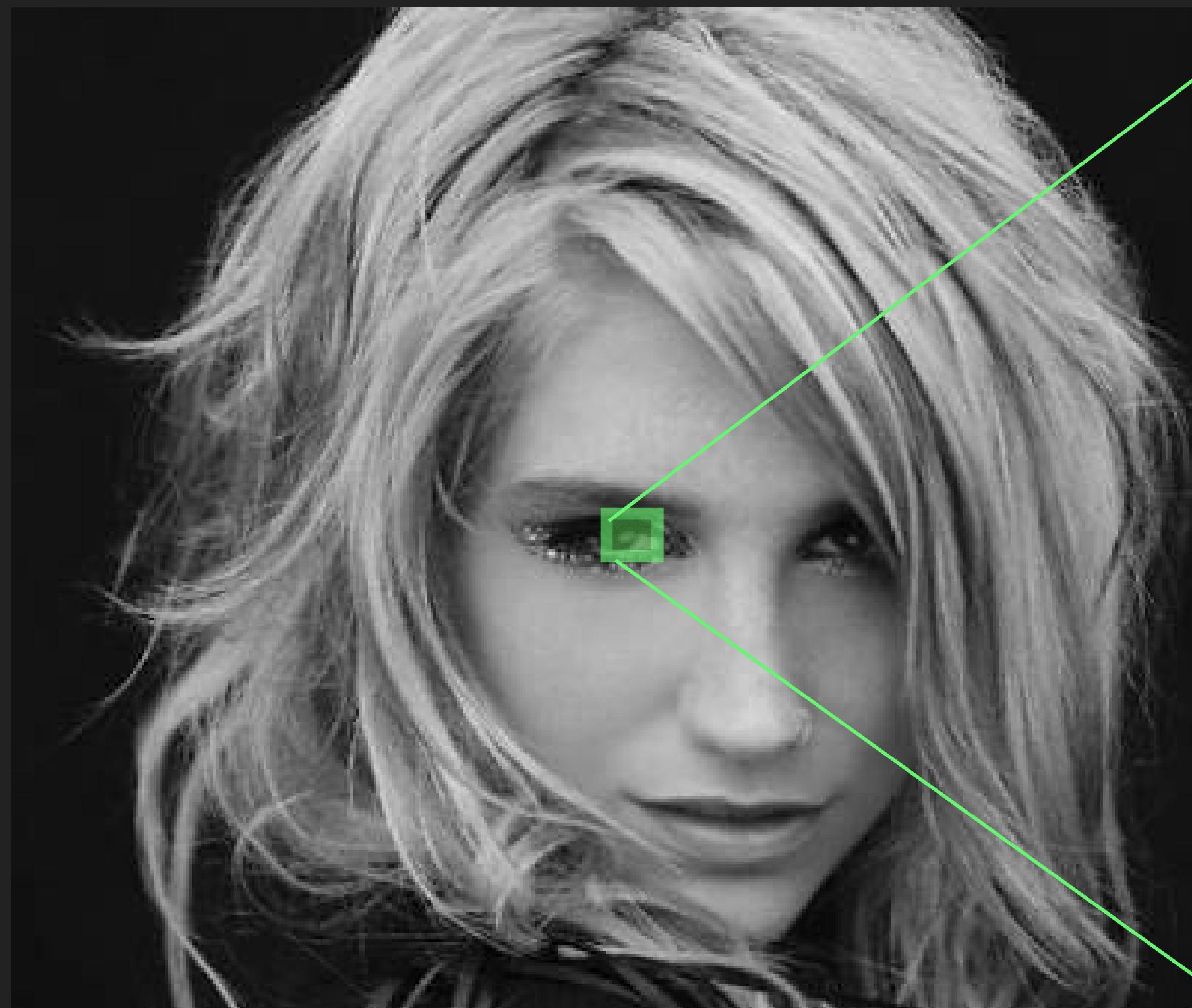
## MASKS - USEFUL FOR IDENTIFYING REGIONS IN AN IMAGE

- ▶ White pixels (1's): Kesha's head
- ▶ Black pixels (0's): not Kesha's head



# GRAYSCALE IMAGE

- ▶ One intensity value per pixel
- ▶ Value indicates: like a shade of gray



8 BIT IMAGE: 256 SHADES OF GRAY (0-255)

Pixel Region (Image Tool 1)

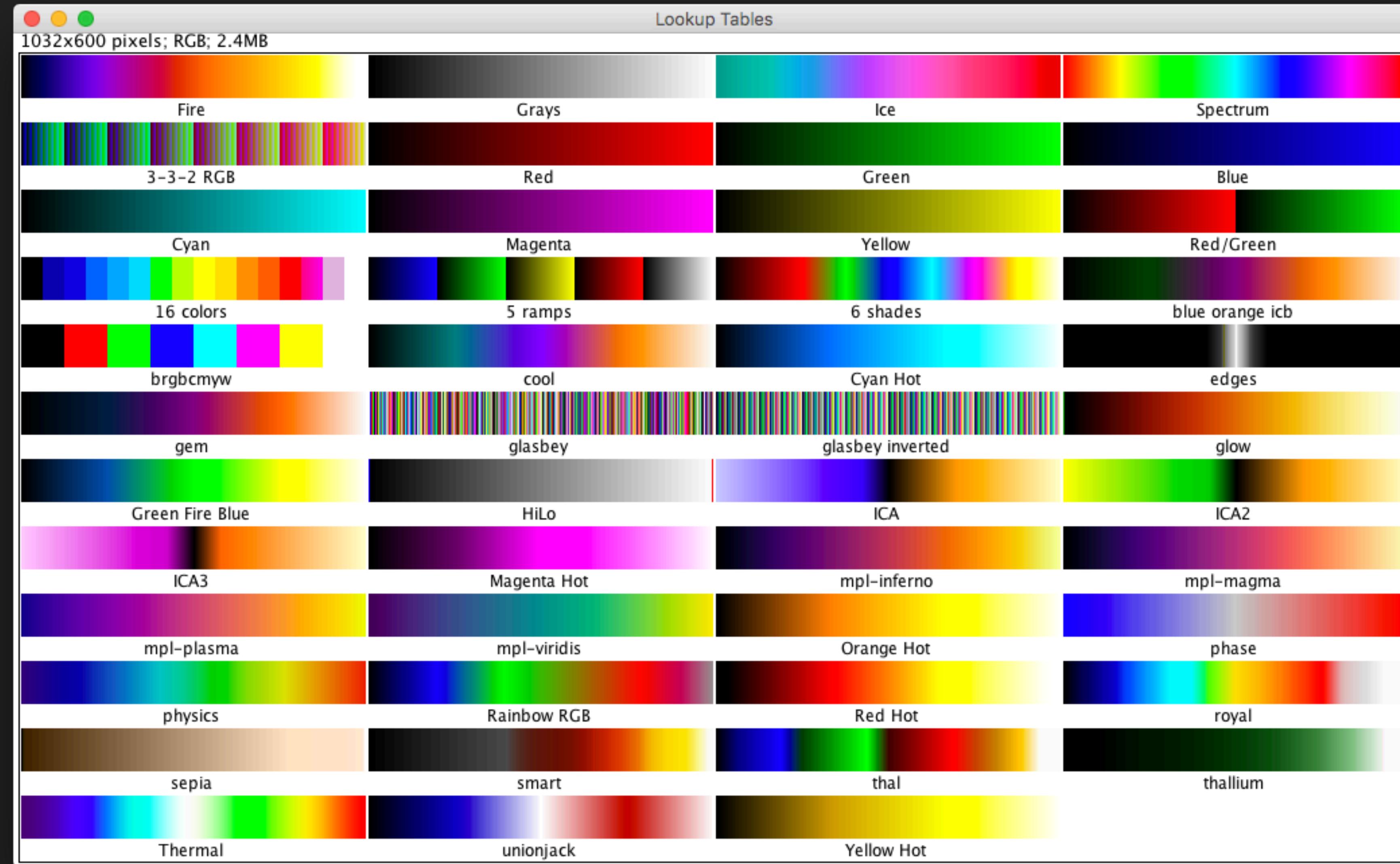
File Edit Window Help

Pixel info: (252, 210) 16

|    |     |     |     |     |     |     |     |     |     |     |     |
|----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| 16 | 12  | 12  | 12  | 16  | 18  | 17  | 19  | 19  | 19  | 17  | 13  |
| 16 | 14  | 16  | 18  | 23  | 29  | 26  | 22  | 19  | 18  | 25  | 32  |
| 6  | 21  | 33  | 46  | 62  | 81  | 88  | 59  | 41  | 29  | 36  | 48  |
| 24 | 54  | 75  | 94  | 103 | 104 | 97  | 77  | 75  | 85  | 114 | 132 |
| 25 | 64  | 91  | 116 | 123 | 118 | 94  | 80  | 85  | 102 | 135 | 158 |
| 26 | 59  | 81  | 100 | 103 | 97  | 72  | 83  | 101 | 127 | 157 | 170 |
| 28 | 49  | 62  | 71  | 69  | 65  | 53  | 90  | 120 | 153 | 174 | 175 |
| 32 | 39  | 32  | 22  | 16  | 21  | 62  | 120 | 155 | 184 | 189 | 177 |
| 31 | 33  | 31  | 29  | 36  | 61  | 128 | 162 | 173 | 175 | 161 | 156 |
| 49 | 61  | 67  | 74  | 86  | 106 | 146 | 156 | 157 | 154 | 144 | 137 |
| 88 | 102 | 107 | 109 | 103 | 101 | 113 | 112 | 109 | 107 | 107 | 106 |

## GRAY SCALE IMAGES

# LOOK-UP TABLES (LUT)



# LOOK-UP TABLES

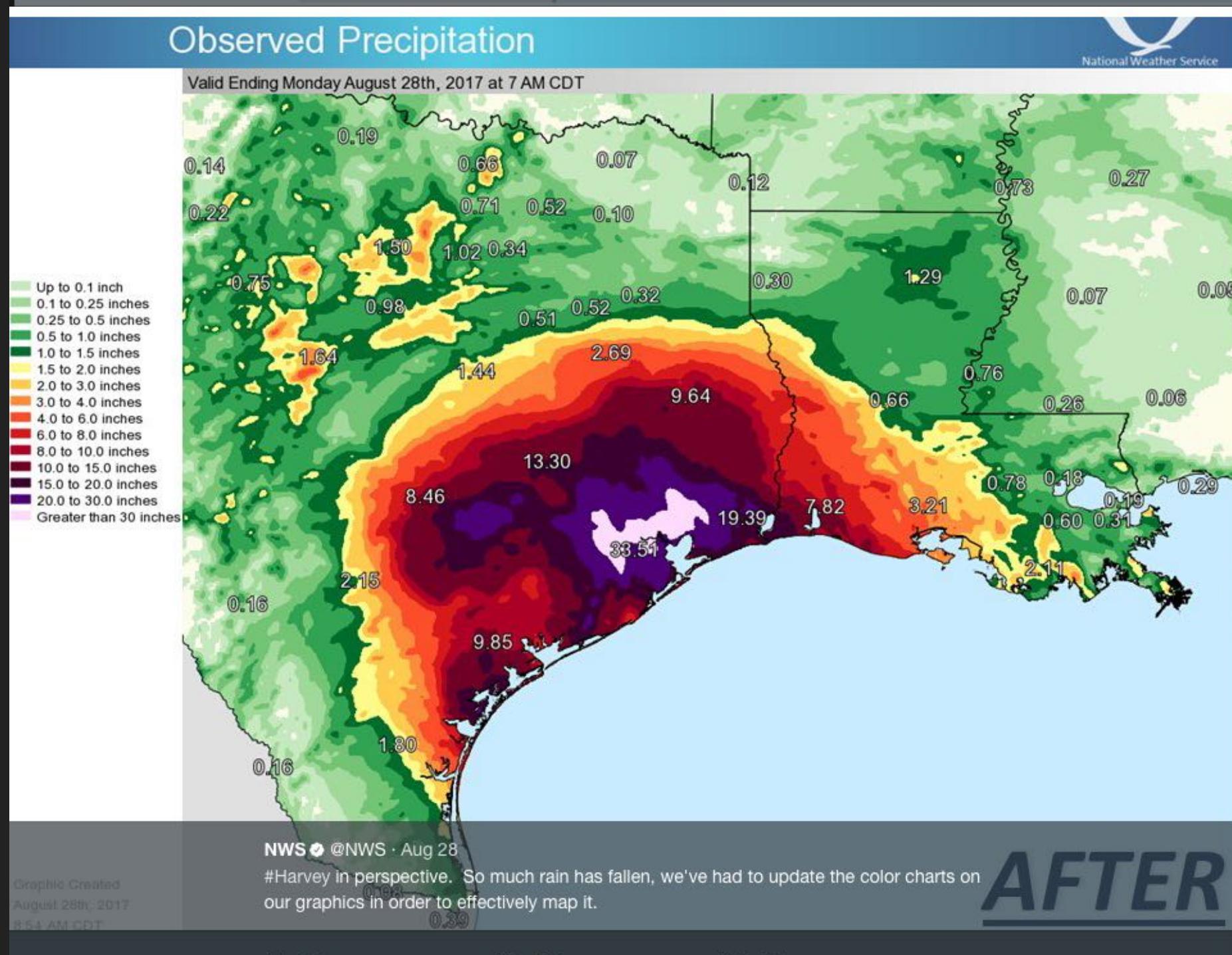
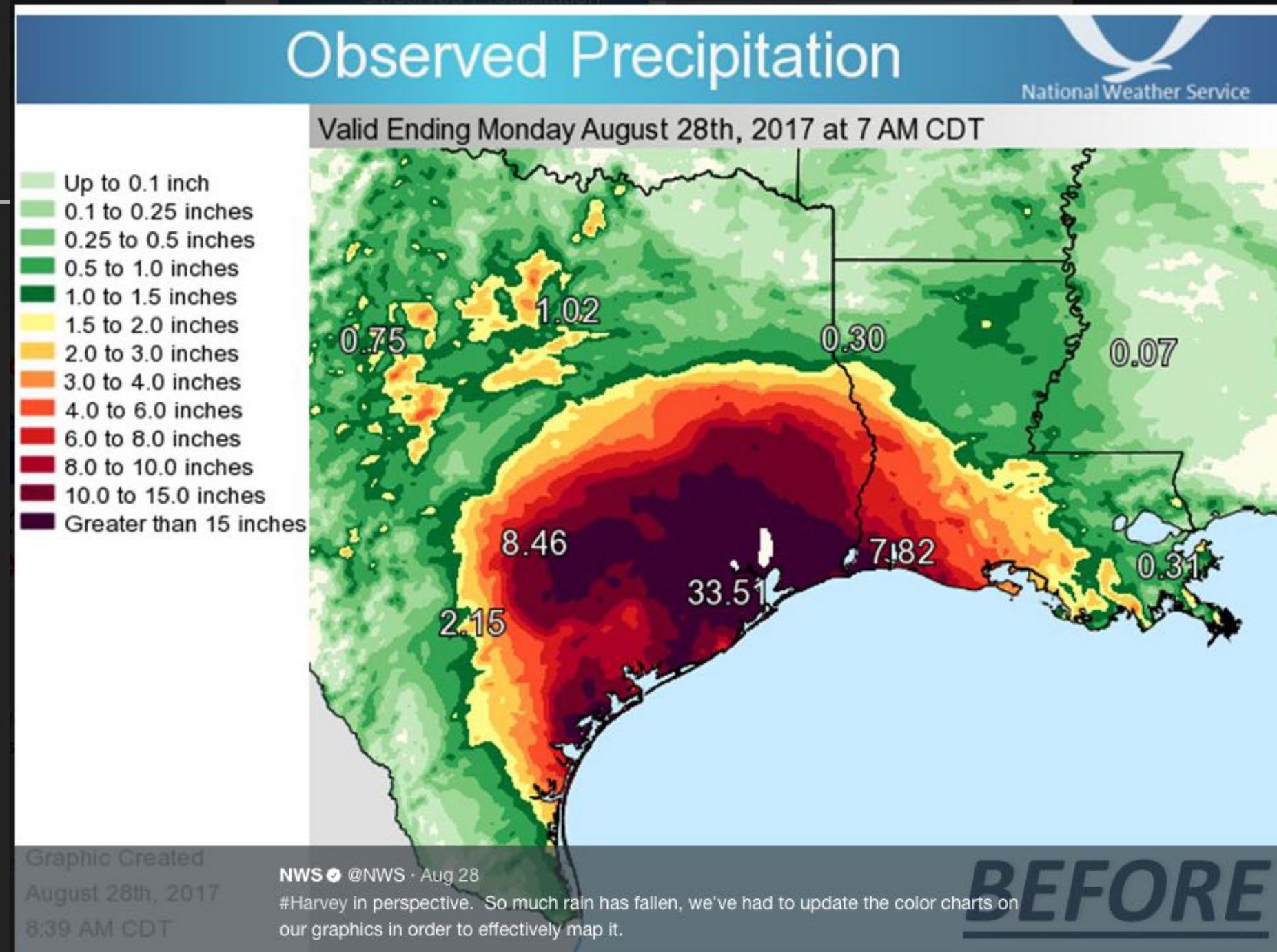


- ▶ These three images are all the *same* grayscale images
- ▶ They are just displayed using three different lookup tables
- ▶ You can see the color mapped to the intensity value in the color bar next to the image

# GRAYSCALE IMAGE

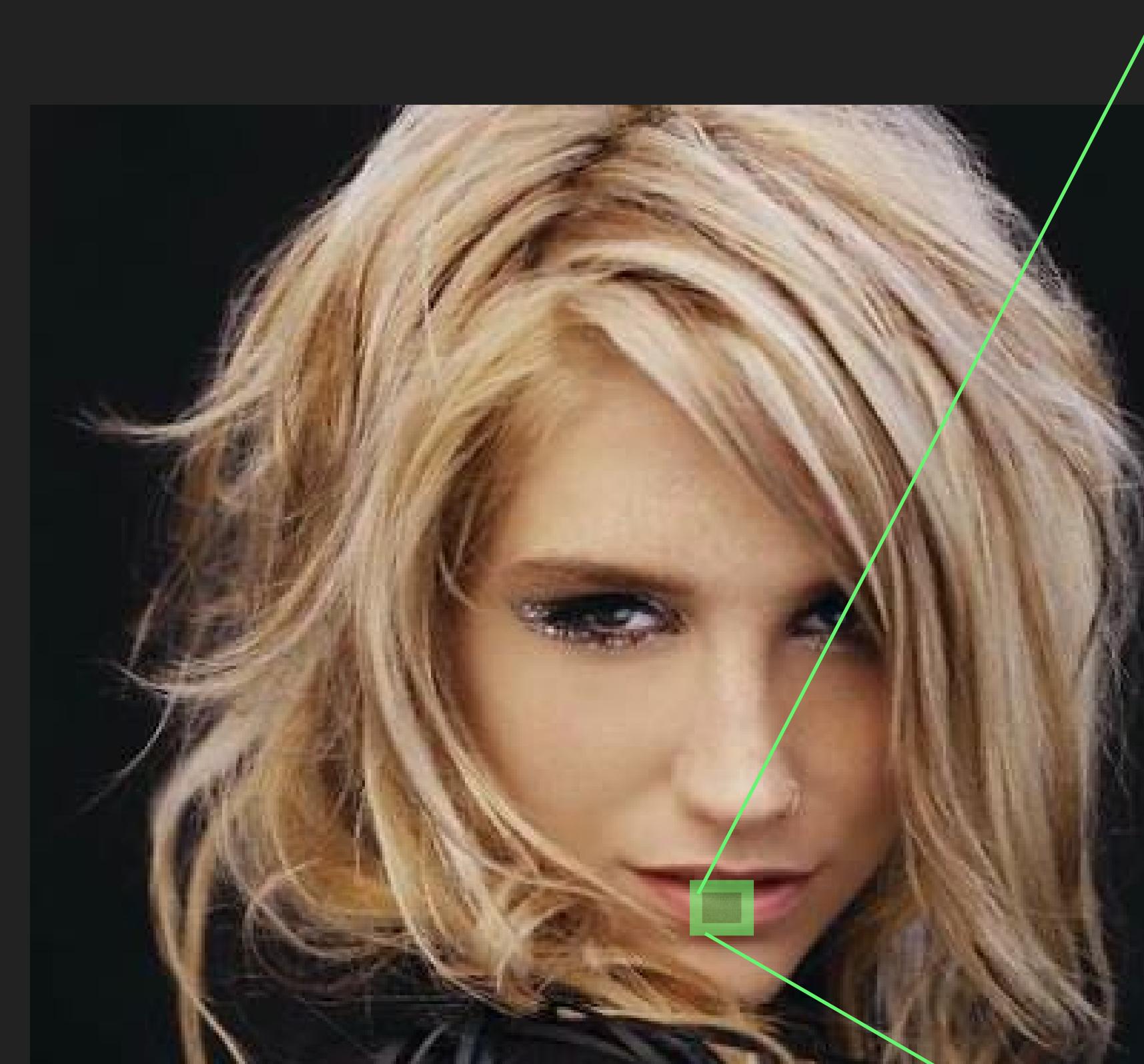
# WEATHER MAPS

- ▶ National Weather Service added New Colors in 2017 so it could Map Harvey's Rains
  - ▶ So much rain fell, they had to update their color charts in their graphics in order to effectively map it.



## RGB IMAGES (AKA TRUECOLOR IMAGES)

- ▶ Each Pixel contains three values used to encode color



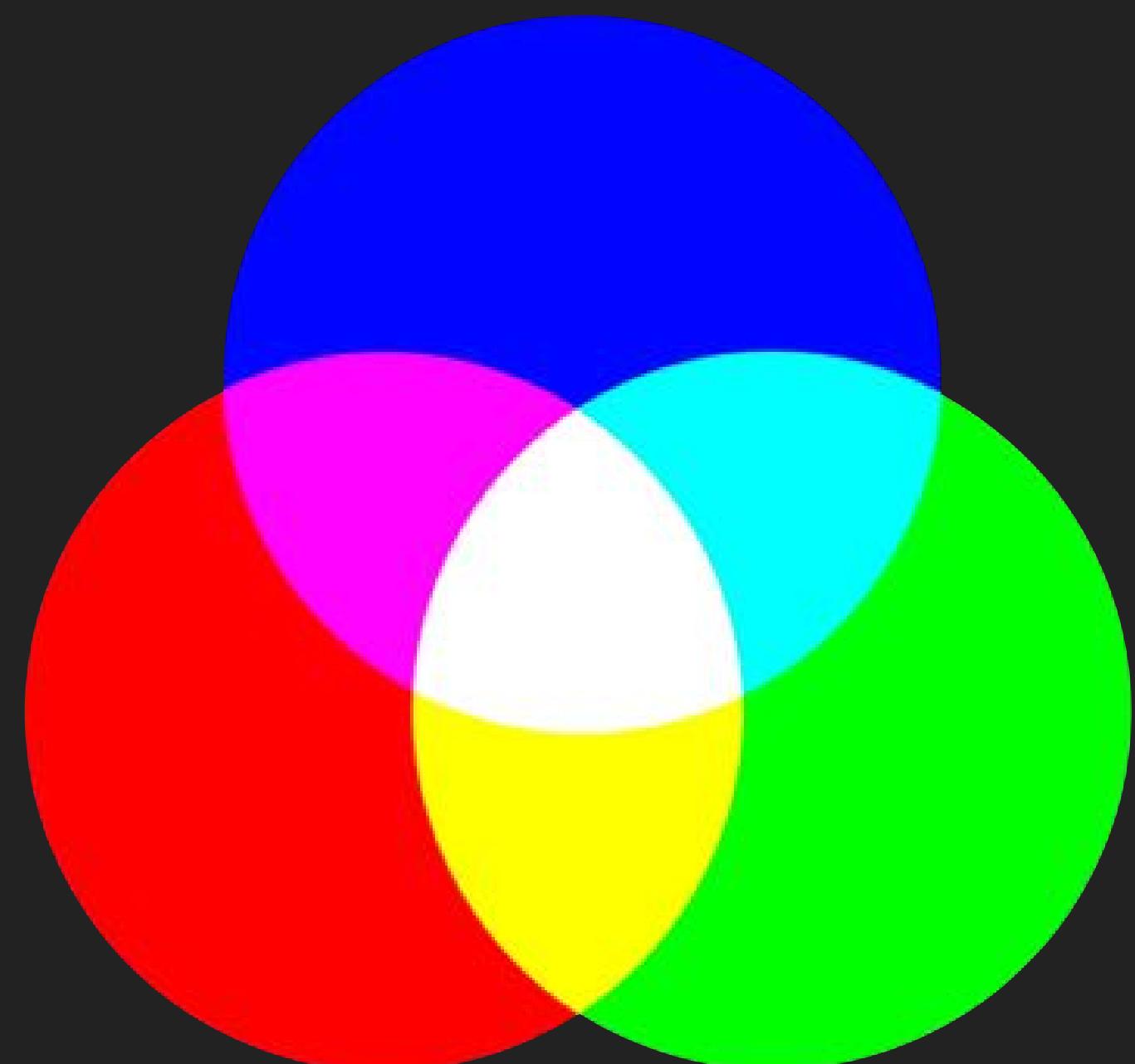
A close-up photograph of a woman with blonde hair, looking directly at the camera. A green square highlighter marks a specific pixel on her cheek, which is then magnified in the bottom right corner of the slide.

|                            |                            |                            |                            |                            |                            |                            |                            |                            |                          |
|----------------------------|----------------------------|----------------------------|----------------------------|----------------------------|----------------------------|----------------------------|----------------------------|----------------------------|--------------------------|
| R: 69<br>G: 10<br>B: 7     | R: 69<br>G: 11<br>B: 7     | R: 71<br>G: 13<br>B: 8     | R: 71<br>G: 14<br>B: 12    | R: 70<br>G: 14<br>B: 11    | R: 70<br>G: 15<br>B: 12    | R: 71<br>G: 17<br>B: 14    | R: 71<br>G: 19<br>B: 15    | R: 73<br>G: 22<br>B: 18    | R: 78<br>G: 27<br>B: 24  |
| R: 83<br>G: 22<br>B: 17    | R: 84<br>G: 23<br>B: 18    | R: 86<br>G: 24<br>B: 22    | R: 86<br>G: 27<br>B: 23    | R: 86<br>G: 26<br>B: 24    | R: 86<br>G: 28<br>B: 26    | R: 88<br>G: 31<br>B: 29    | R: 91<br>G: 32<br>B: 31    | R: 94<br>G: 38<br>B: 36    | R: 101<br>G: 45<br>B: 44 |
| R: 107<br>G: 44<br>B: 39   | R: 110<br>G: 47<br>B: 41   | R: 111<br>G: 48<br>B: 44   | R: 112<br>G: 50<br>B: 45   | R: 107<br>G: 47<br>B: 43   | R: 110<br>G: 48<br>B: 46   | R: 113<br>G: 53<br>B: 51   | R: 117<br>G: 56<br>B: 55   | R: 122<br>G: 63<br>B: 62   | R: 129<br>G: 69<br>B: 68 |
| R: 127<br>G: 62<br>B: 57   | R: 130<br>G: 65<br>B: 62   | R: 135<br>G: 70<br>B: 65   | R: 136<br>G: 71<br>B: 68   | R: 134<br>G: 69<br>B: 66   | R: 136<br>G: 71<br>B: 70   | R: 141<br>G: 77<br>B: 76   | R: 144<br>G: 80<br>B: 80   | R: 149<br>G: 85<br>B: 85   | R: 151<br>G: 87<br>B: 87 |
| R: 143<br>G: 77<br>B: 71   | R: 149<br>G: 81<br>B: 75   | R: 153<br>G: 86<br>B: 82   | R: 157<br>G: 88<br>B: 86   | R: 160<br>G: 91<br>B: 90   | R: 163<br>G: 93<br>B: 93   | R: 167<br>G: 96<br>B: 96   | R: 169<br>G: 100<br>B: 100 | R: 170<br>G: 100<br>B: 102 | R: 166<br>G: 97<br>B: 98 |
| R: 172<br>G: 103<br>B: 98  | R: 175<br>G: 103<br>B: 100 | R: 177<br>G: 107<br>B: 104 | R: 180<br>G: 109<br>B: 107 | R: 183<br>G: 109<br>B: 110 | R: 183<br>G: 109<br>B: 110 | R: 183<br>G: 109<br>B: 110 | R: 182<br>G: 108<br>B: 109 | R: 178<br>G: 103<br>B: 106 | R: 171<br>G: 95<br>B: 99 |
| R: 200<br>G: 129<br>B: 125 | R: 200<br>G: 126<br>B: 122 | R: 197<br>G: 123<br>B: 120 | R: 196<br>G: 122<br>B: 120 | R: 192<br>G: 117<br>B: 116 | R: 188<br>G: 112<br>B: 112 | R: 184<br>G: 104<br>B: 112 | R: 177<br>G: 99<br>B: 107  | R: 173<br>G: 94<br>B: 102  | R: 166<br>G: 87<br>B: 96 |
| R: 211<br>G: 137<br>B: 134 | R: 207<br>G: 133<br>B: 129 | R: 201<br>G: 127<br>B: 124 | R: 198<br>G: 122<br>B: 120 | R: 193<br>G: 116<br>B: 115 | R: 186<br>G: 108<br>B: 108 | R: 177<br>G: 97<br>B: 99   | R: 168<br>G: 88<br>B: 91   | R: 161<br>G: 81<br>B: 85   | R: 157<br>G: 77<br>B: 81 |
| R: 202<br>G: 128<br>B: 122 | R: 196<br>G: 120<br>B: 116 | R: 188<br>G: 113<br>B: 109 | R: 182<br>G: 107<br>B: 102 | R: 178<br>G: 101<br>B: 96  | R: 176<br>G: 97<br>B: 99   | R: 170<br>G: 93<br>B: 99   | R: 166<br>G: 88<br>B: 92   | R: 161<br>G: 83<br>B: 92   | R: 159<br>G: 80<br>B: 90 |

# COLOR

## RGB IMAGES

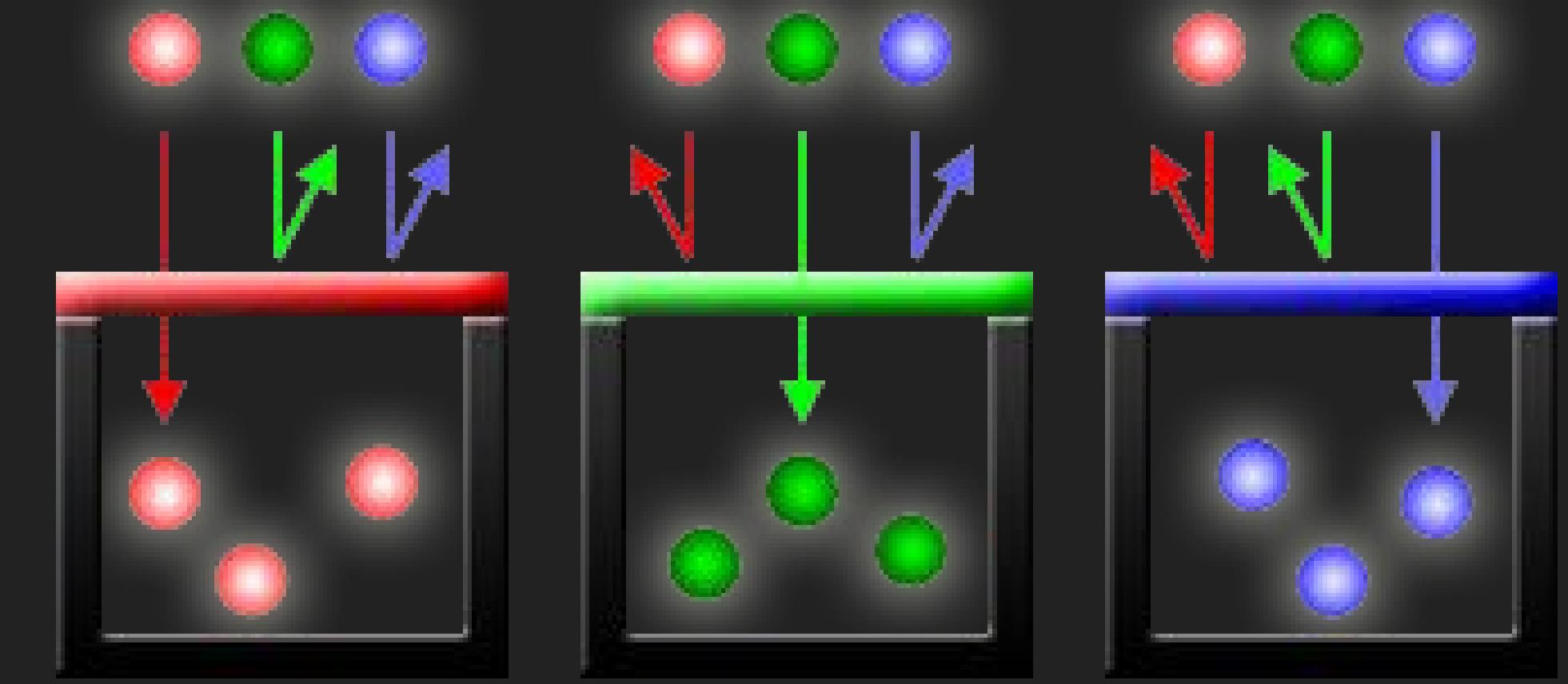
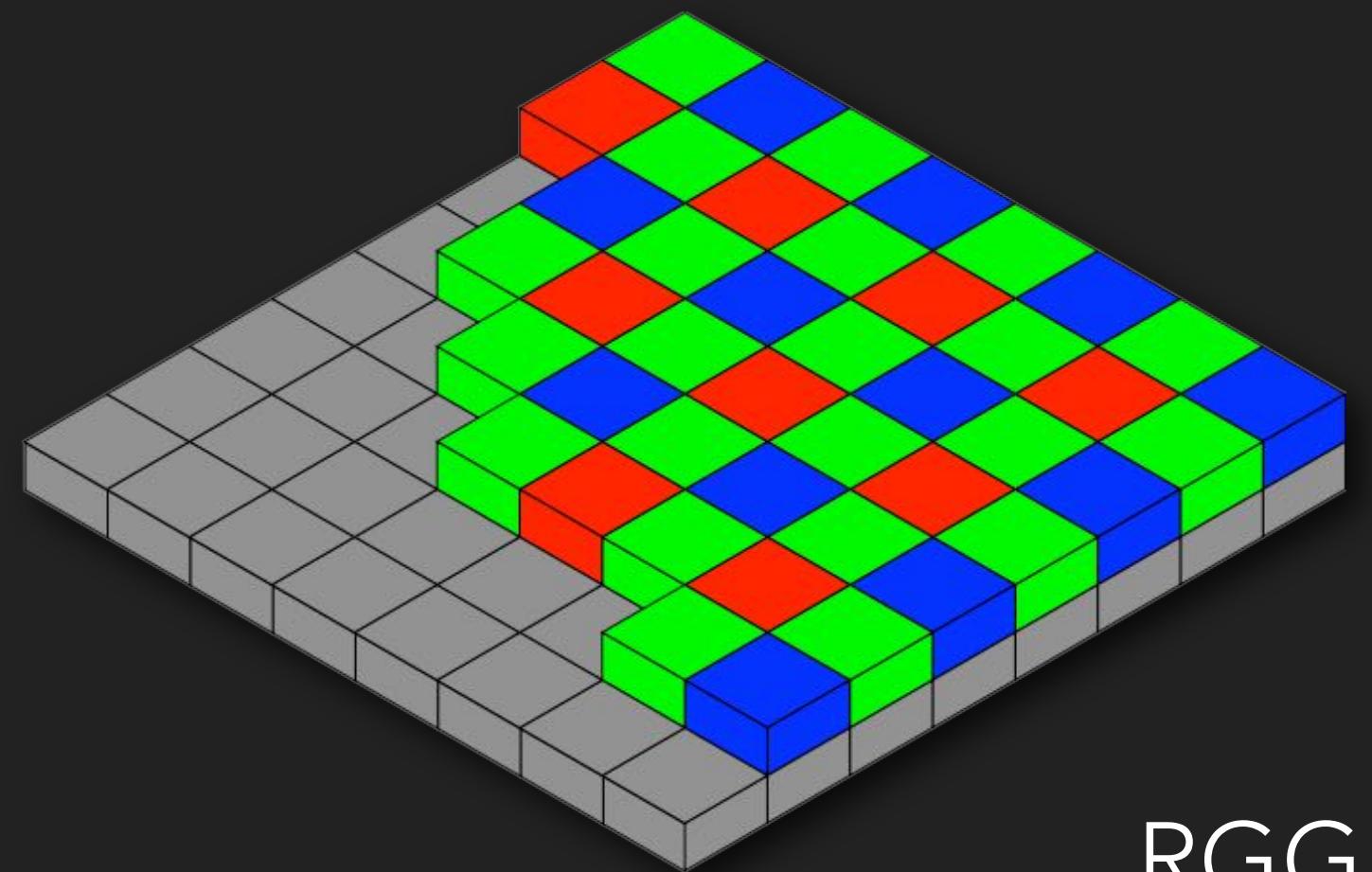
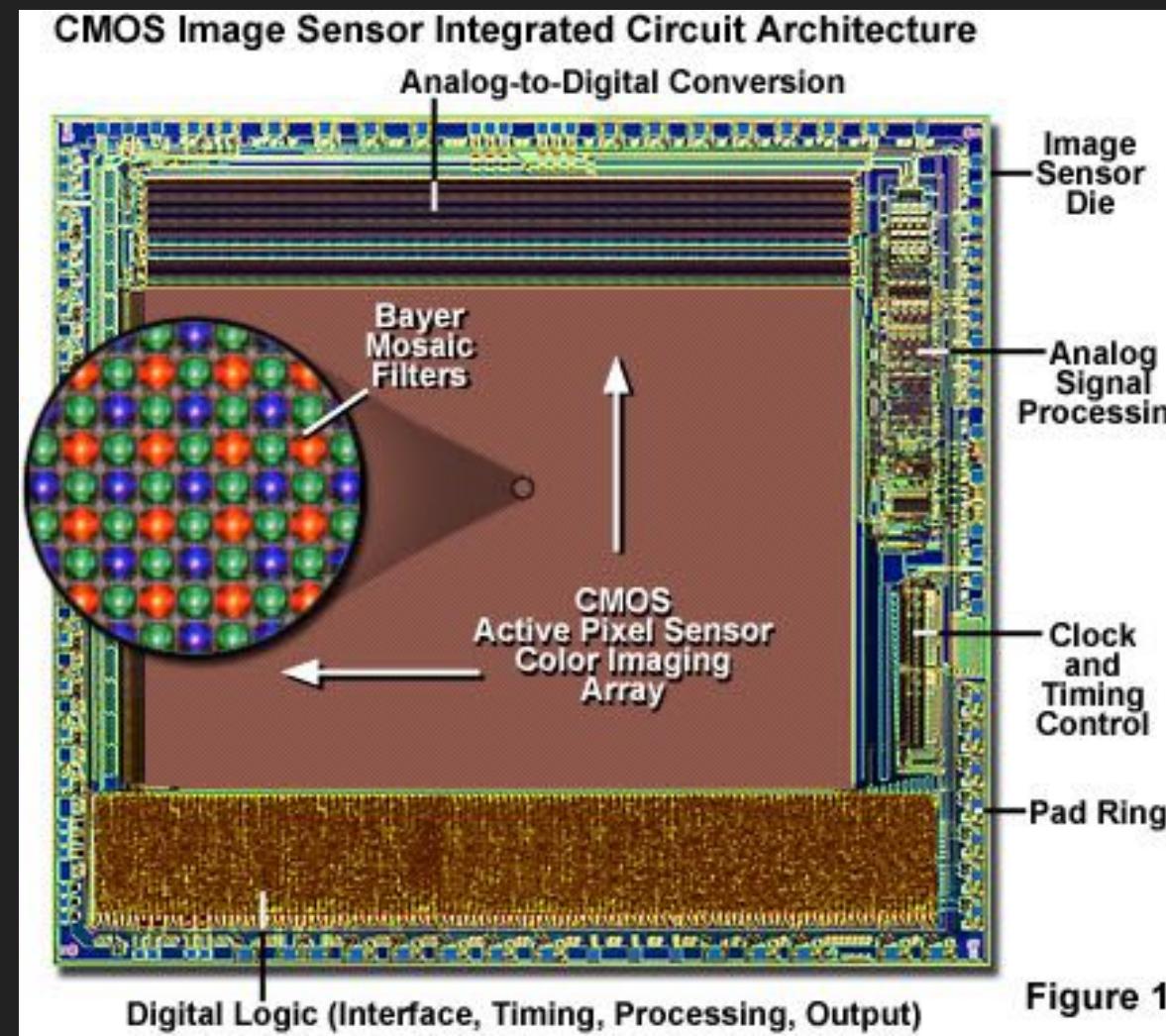
- ▶ Each pixel has a triplet of intensity values, one for each channel (Red,Green,Blue)
- ▶ Color of a pixel is determined by the combination of the values of the 3 channels
- ▶ Based on Color Theory - similar to the cone sensitivities in our eye



| INTENSITY RANGE PER CHANNEL |                         |                         |                         |                         |                         |
|-----------------------------|-------------------------|-------------------------|-------------------------|-------------------------|-------------------------|
| Red: 0 -255                 |                         |                         |                         |                         |                         |
| Green: 0-255                |                         |                         |                         |                         |                         |
| Blue: 0-255                 |                         |                         |                         |                         |                         |
| R: 58<br>G:180<br>B: 93     | R: 49<br>G:191<br>B:119 | R: 42<br>G:154<br>B:145 | R: 50<br>G:133<br>B:154 | R: 45<br>G:151<br>B:214 | R: 36<br>G:190<br>B:228 |
| R: 44<br>G:133<br>B:157     | R: 44<br>G:156<br>B:115 | R: 50<br>G:152<br>B:112 | R: 46<br>G:105<br>B:153 | R: 43<br>G:123<br>B:237 | R: 45<br>G:126<br>B:217 |
| R: 51<br>G: 86<br>B:187     | R: 34<br>G:135<br>B:172 | R: 37<br>G:130<br>B:138 | R: 40<br>G: 83<br>B:162 | R: 47<br>G: 73<br>B:184 | R: 47<br>G: 59<br>B:221 |
| R: 37<br>G: 43<br>B:221     | R: 33<br>G: 41<br>B:255 | R: 37<br>G: 43<br>B:165 | R: 53<br>G: 37<br>B:153 | R: 60<br>G: 39<br>B:166 | R: 58<br>G: 34<br>B:241 |
| R: 40<br>G: 25<br>B:163     | R: 37<br>G: 31<br>B:182 | R: 65<br>G: 38<br>B:114 | R: 79<br>G: 61<br>B: 91 | R:118<br>G: 85<br>B:110 | R:122<br>G: 51<br>B:173 |
| R: 39<br>G: 26<br>B:172     | R: 43<br>G: 33<br>B:130 | R: 86<br>G: 60<br>B: 99 | R:179<br>G:101<br>B: 85 | R:209<br>G:117<br>B: 82 | R:221<br>G:107<br>B:101 |
| R: 42                       | R: 50                   | R:109                   | R:255                   | R:255                   | R:251                   |

## COLOR MODE

# IMAGE SENSORS THAT CAPTURE RGB HAVE A COLORED FILTER ARRAY ON THEM



RGGB

- ▶ Bayer Array - alternating rows of red, green, and blue filters
- ▶ Extra green cells - humans are more sensitive to green light
- ▶ **Contrast of image Reduced** by a third or a fourth (because you have to interpolate the color values)
- ▶ Fluorescent Microscope cameras do not have a Bayer filter: better contrast