



# Microscope design 1.7

## SC865

Concepts for maximum image contrast



# § 1 Paraxial Optics

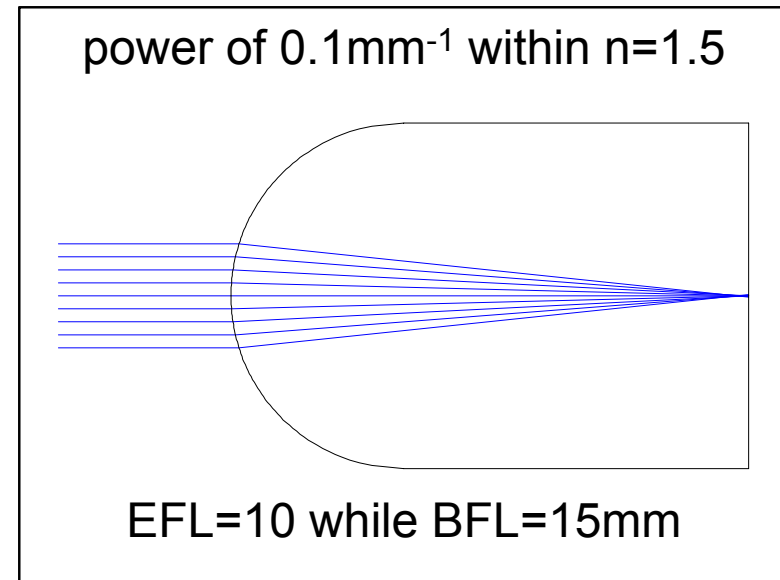
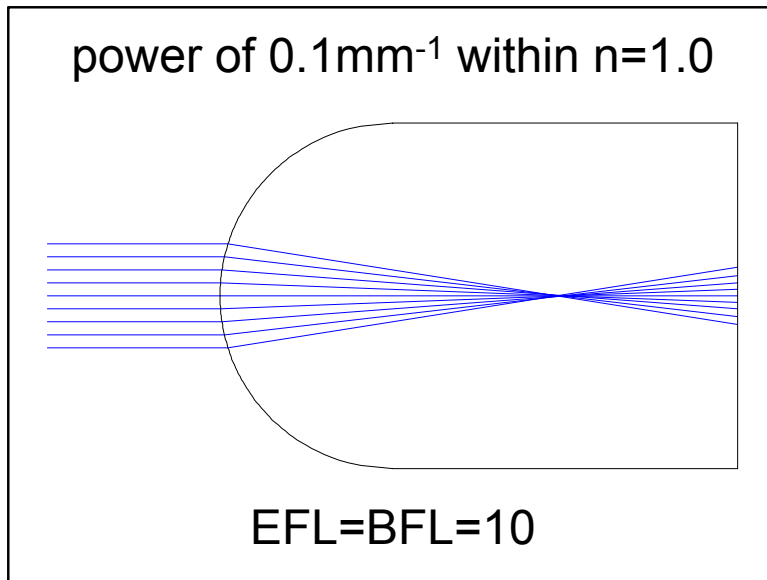
Perfect lens elements  
for  
Optical specifications



# Focal length and effective focal length

$$f_n = nf$$
$$\sin \theta_n = \frac{\sin \theta}{n}$$

- Effective focal length EFL=f
  - Constant throughout all media
- Focal length FL= $f_n$ 
  - Scales with refractive index
- Due to Snell's Law



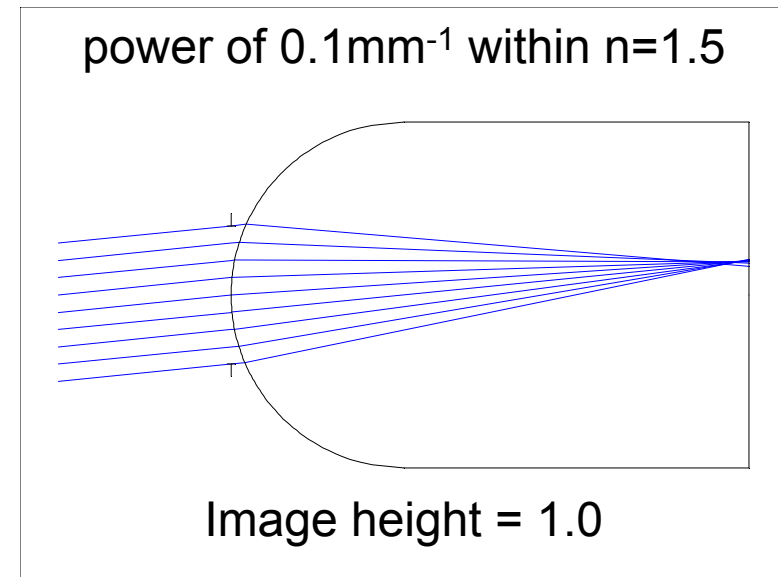
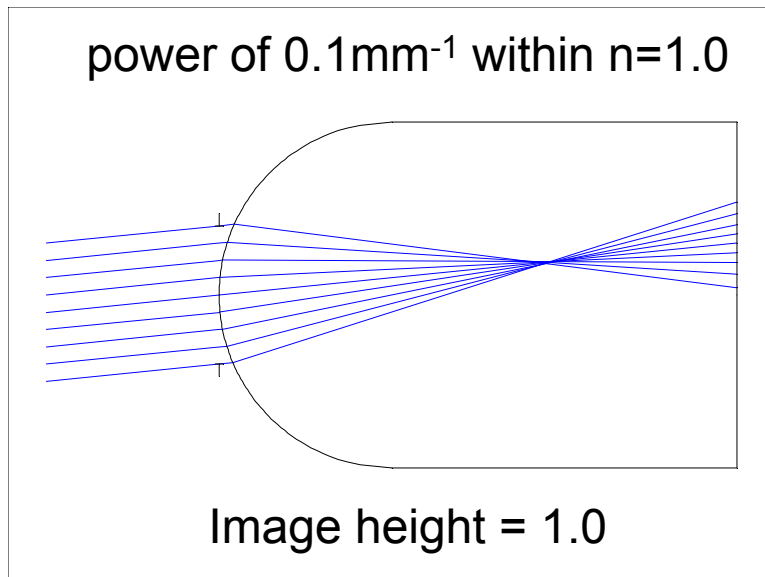


# Image height

$$h_i = \theta_0 f_0$$

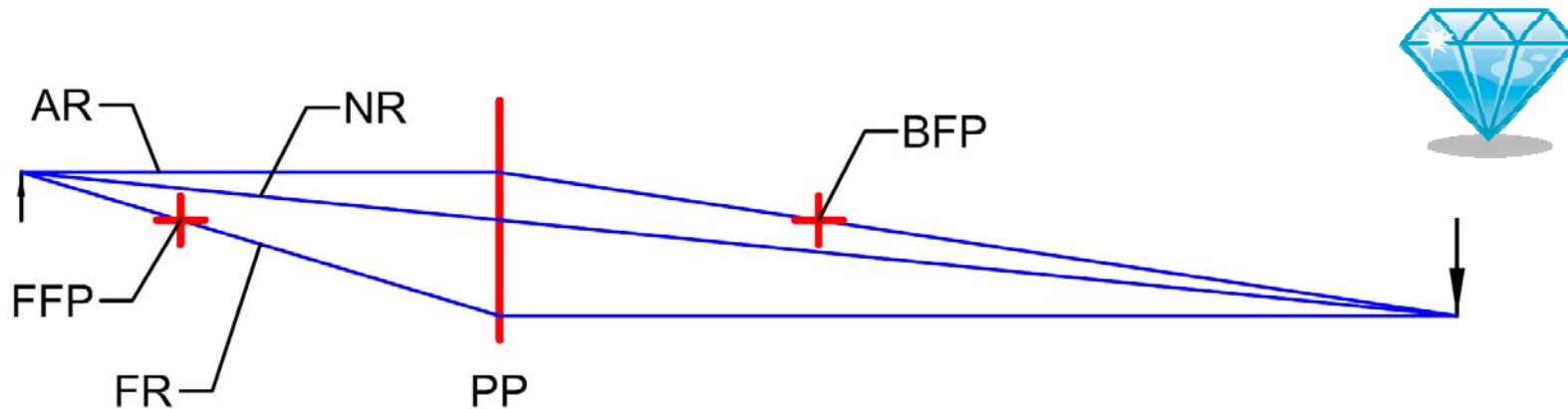
$$h_i = \theta_n f_n$$

- Constant with EFL
- Decedent upon
  - Object height
  - EFL





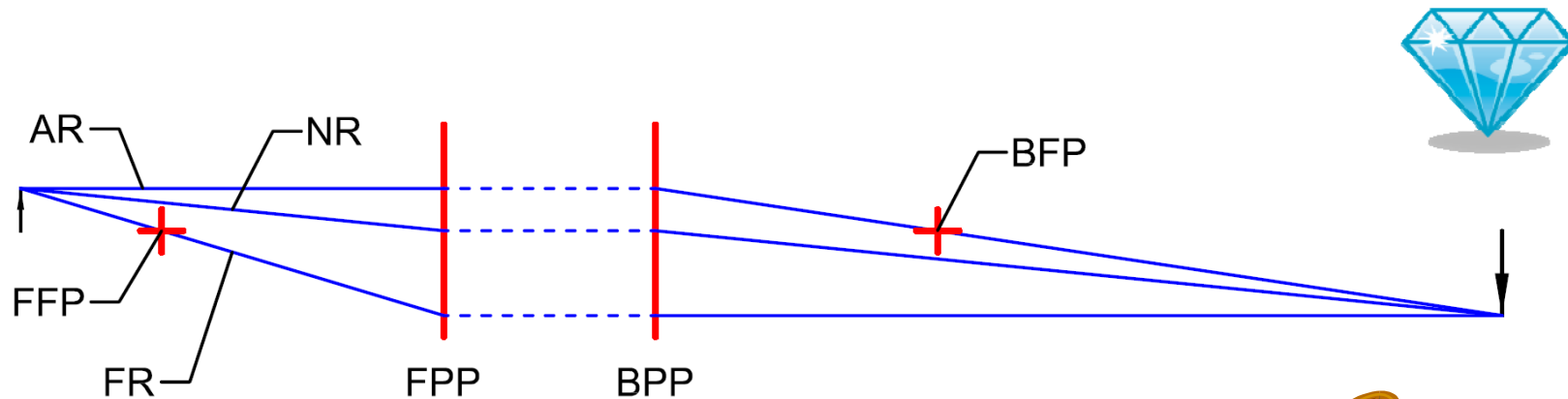
# Thin lens cardinal points



- Object cardinal points
  - Front focal point FFP
  - Principle plane PP
- Object Rays
  - Axial ray AR parallel to axis
  - Nodal ray NR thru PP center
  - Focal ray FR thru FFP
- Image cardinal points
  - Back focal point BFP
  - Principle plane PP
- Image Rays
  - AR directed thru BFP
  - NR passes without bend
  - FR becomes axial



# Thick lens separates PP



- Principle planes
  - Front principle plane FPP
  - Back principle plane BPP
  - Transformation between planes at right

$$\begin{vmatrix} y' \\ m' \end{vmatrix} = \begin{vmatrix} 1 & 0 \\ -1/f & 1 \end{vmatrix} \cdot \begin{vmatrix} y \\ m \end{vmatrix}$$

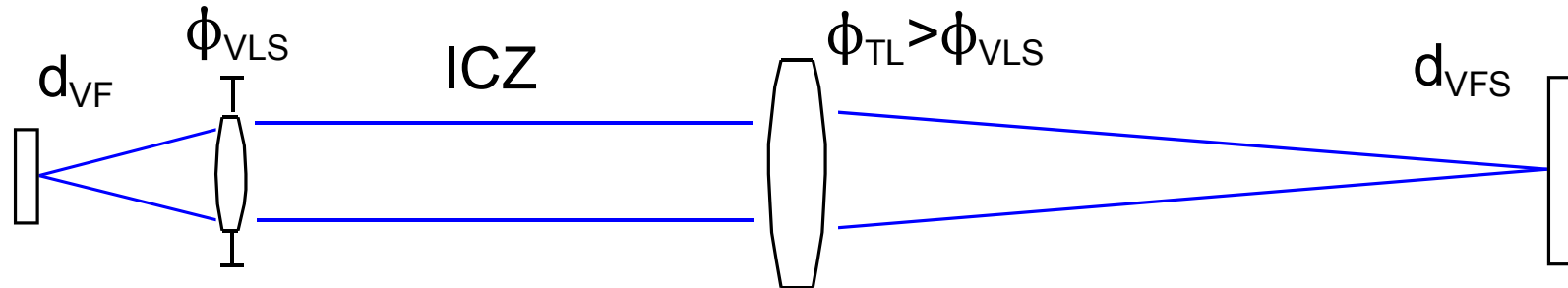


## § 2 Stops

Constraints in space and angle



# Vision stops



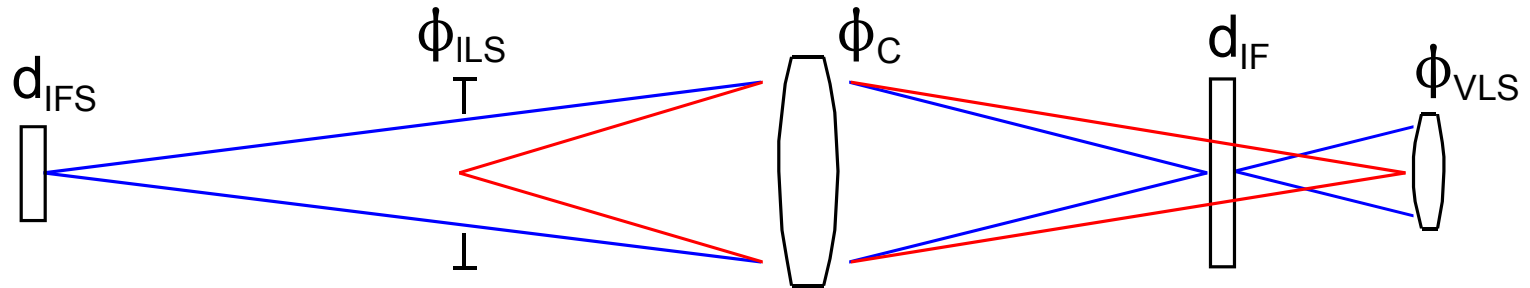
- Spatial extents
  - vision field-stop VFS
  - vision field VF
- Angular extents
  - vision lens-stop VLS
- Image conjugates
  - VF and VFS
- Other features
  - infinity correction zone ICZ
  - tube lens TL







# Illumination stops



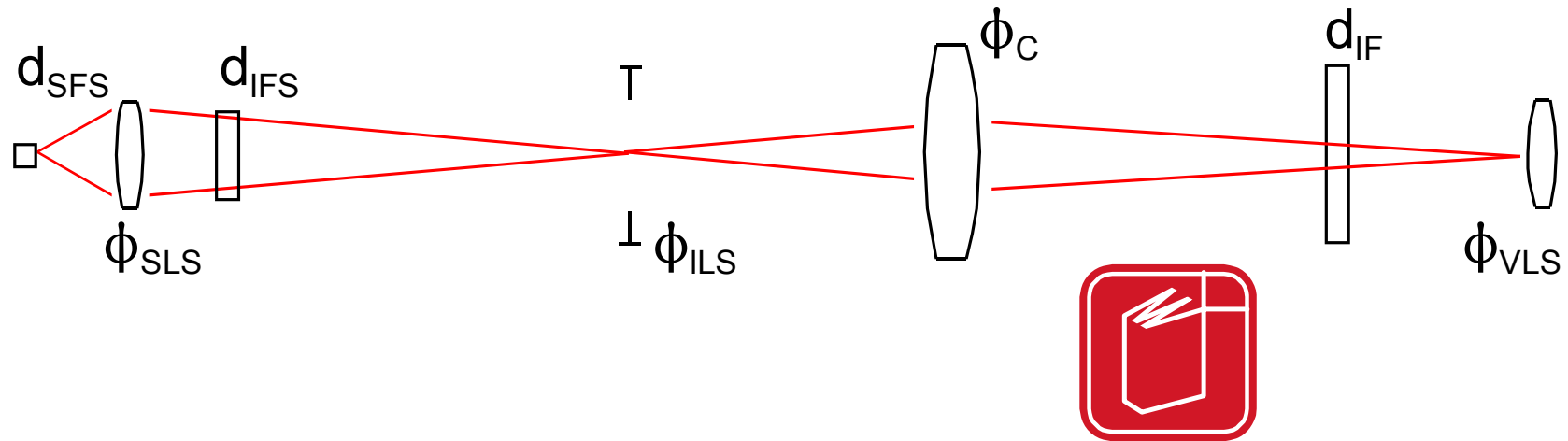
- Spatial extents
  - IFS illumination field-stop
  - IF illumination field
- Angular extents
  - ILS illumination lens-stop

- Image conjugates
  - IFS and IF
  - ILS and VLS
- Other features
  - $\Phi_C$  Diameter of condenser





# Source stops



- Spatial extents
  - source field-stop SFS
- Angular extents
  - source lens-stop SLS
- Source image located at
  - illumination lens-stop ILS
  - vision lens-stop VLS
- Other features
  - Diffuser (not shown)



## § 3 Wave Optics

Propagation of electromagnetic waves



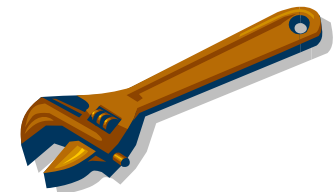
# Space-angle product of waves

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$$A\Omega \geq \lambda^2$$



Cannot be made smaller  
without  
creation of evanescent field



$$A_0\Omega_0 = A_1\Omega_1 = \dots = A_n\Omega_n \geq \lambda^2$$



# Coherence length

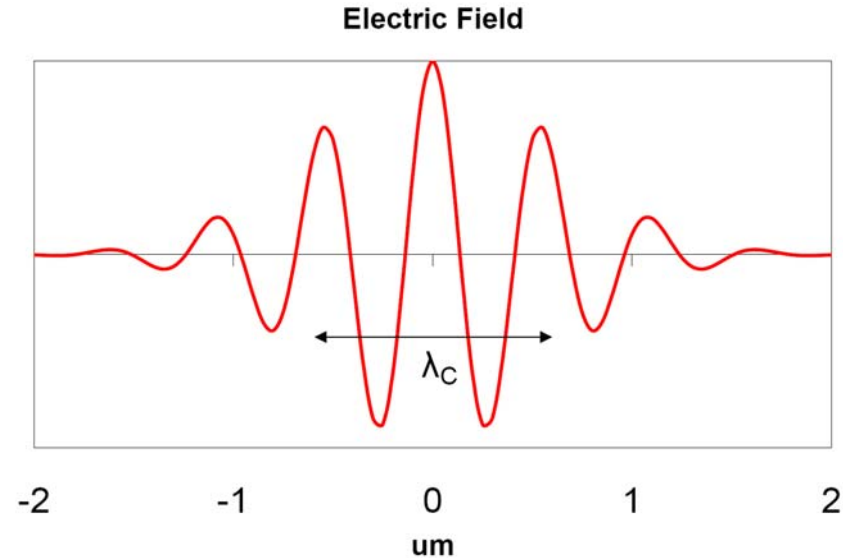
- Analogous to Heisenberg Uncertainty principle
- conversion of  $\sigma_z \sigma_k$  of to wavelength

$$\Delta z \Delta p \geq \frac{\hbar}{2}$$
$$\sigma_z \sigma_k \geq \frac{1}{2}$$



$$\Delta \lambda = 4 \sigma_\lambda$$
$$\lambda_c = 4 \sigma_z$$
$$\lambda_c = \frac{\lambda^2}{\Delta \lambda}$$

- Human vision filter
  - $\lambda = 550\text{nm}$
  - $\Delta \lambda = 650\text{nm} - 400\text{nm}$
  - $\lambda_c = 1.2\mu\text{m} \approx 2\lambda$



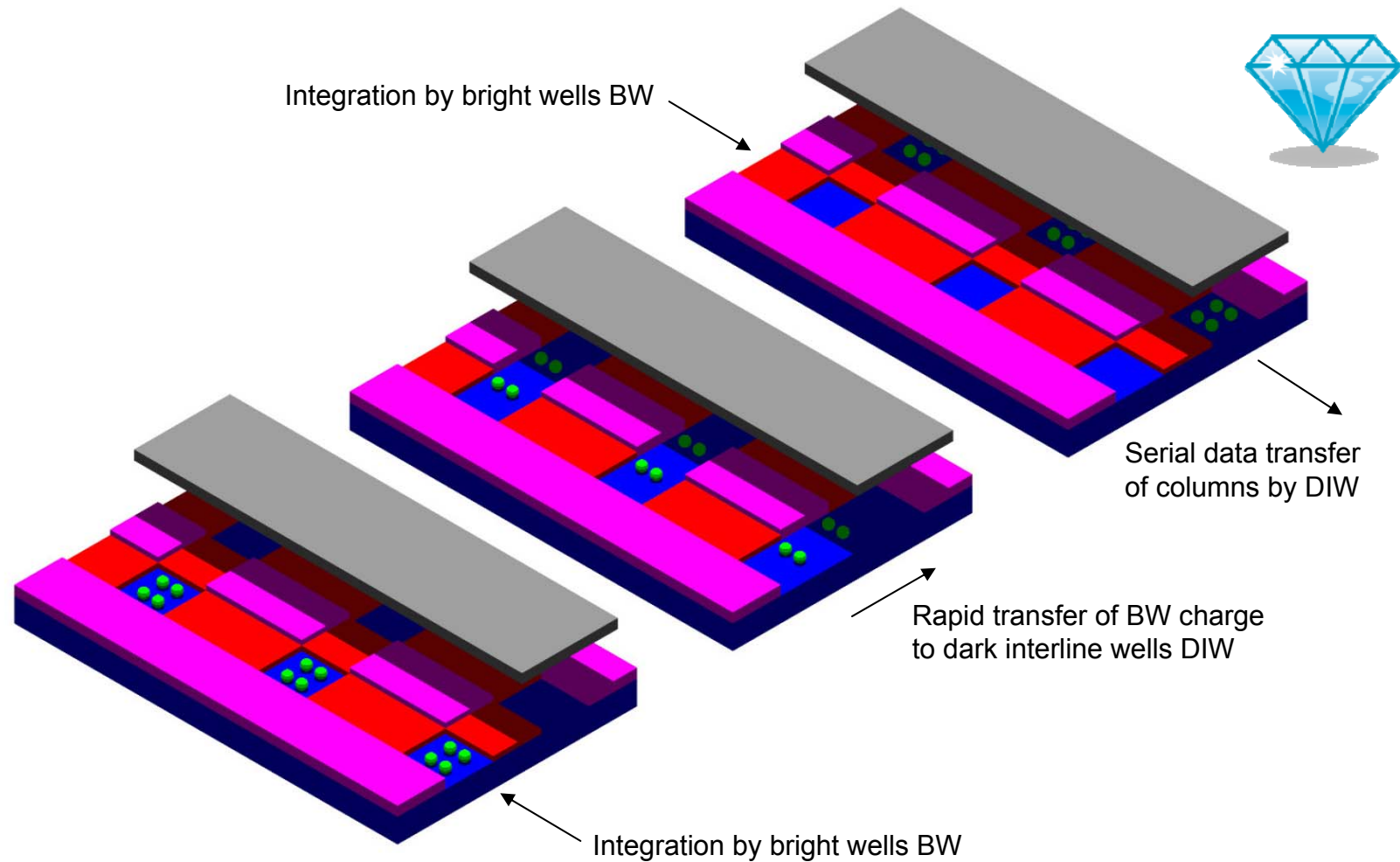


## § 4 Charge coupled devices

**Space-angle product of pixel  
determines  
design goals for optics**



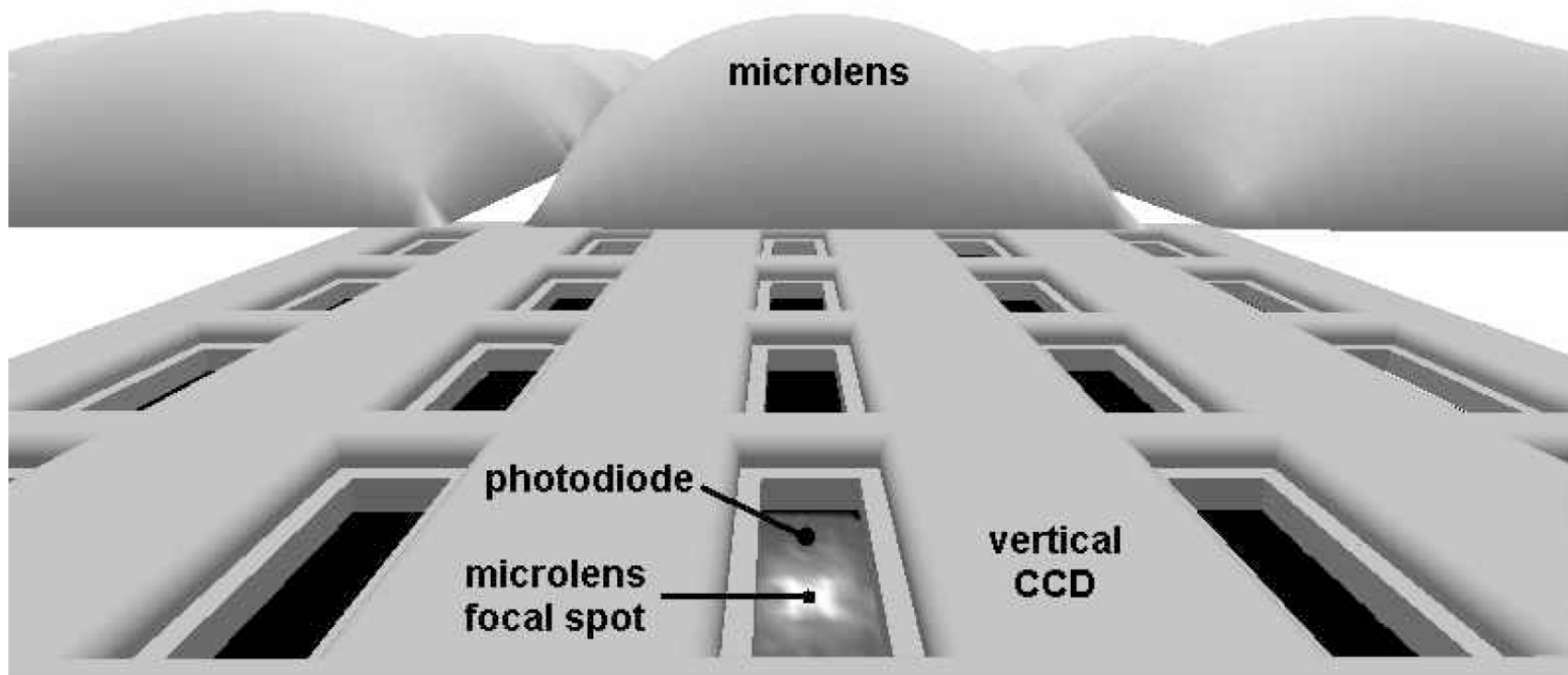
# Interline transfer with shield





# CCD microlens array

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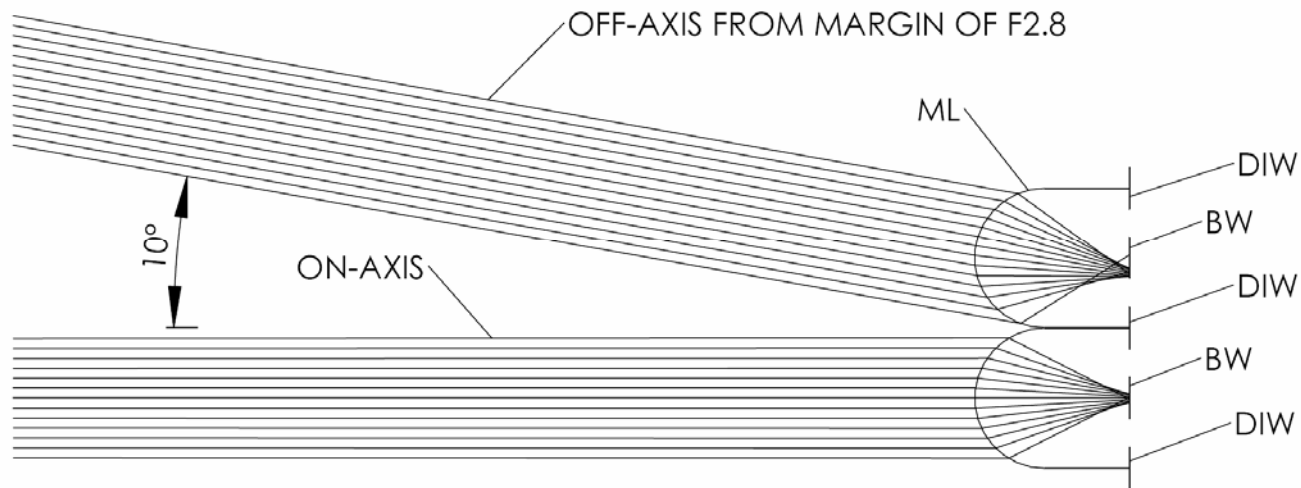
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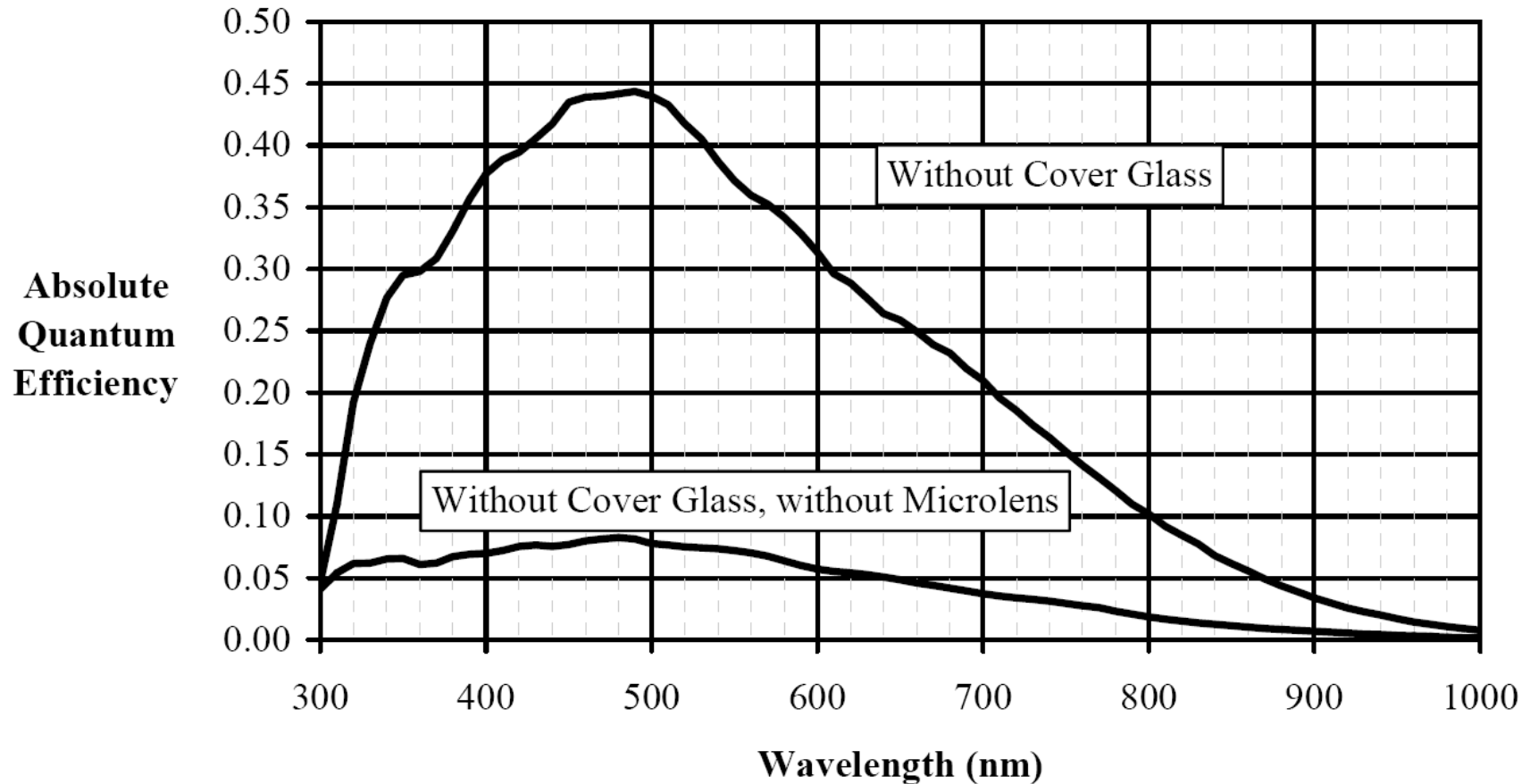
# F2.8 Microlens



- Bright wells BW
  - accumulate charge during exposure
  - display less than 25% fill-factor
- Dark interline wells DIW
  - Quickly collect charge from bright wells
  - Transfer charge without further exposure
- Microlenses ML
  - Display nearly 100% fill-factor
  - Accept F2.8 rays
  - Spherical aberrations SA
  - Surface form errors SFE
  - >4X gain to quantum efficiency



# Quantum efficiency KAI-1020



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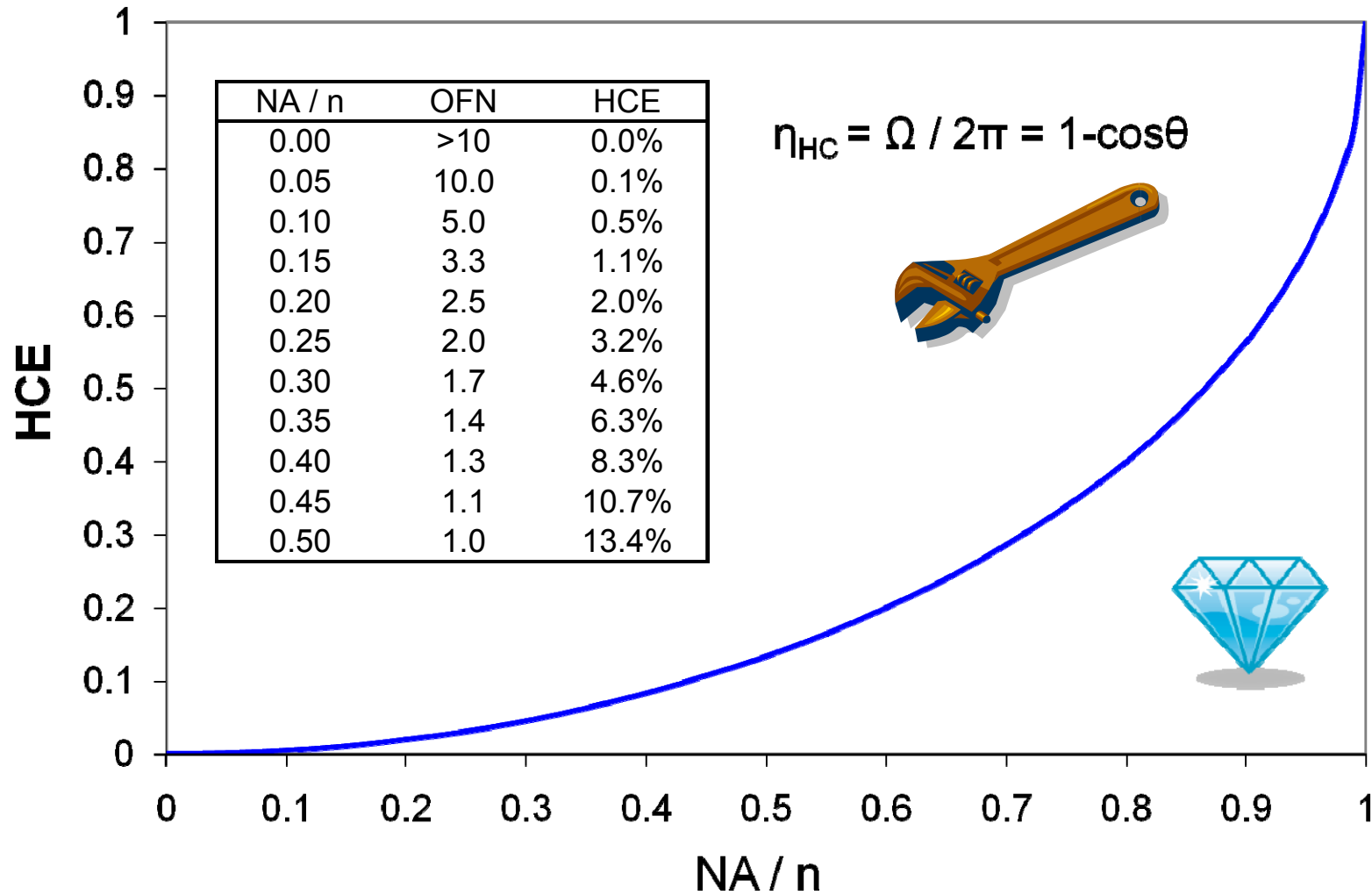


## § 5 Contrast enhancement

To make a needle appear within haystack



# Hemispherical Collection Efficiency $\eta_{\text{HC}}$





# Sub-pixel collection efficiency

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- Angle at object

$$\begin{aligned}\Omega_{pp} &= 4\pi \sin^2 \frac{\theta_{pp}}{2} \\ &\approx \frac{(ONA)^2}{2} H\end{aligned}$$

- Angle at image

$$\begin{aligned}\Omega_{pp} &= 4\pi \sin^2 \frac{\theta_p}{2} \\ &\approx \frac{(INA)^2}{2M^2} H \\ &\approx \frac{H}{8M^2(IFN)^2}\end{aligned}$$

A hemisphere H is reference as

$$H = 2\pi$$



# Full-pixel collection efficiency

- Space-angle product at object
- Space-angle product at image

$$A_{pp}\Omega_{pp} = \frac{d_p^2}{M^2} 4\pi \sin^2 \frac{\theta_{pp}}{2}$$
$$\approx \frac{d_p^2}{M^2} \frac{(ONA)^2}{2} H$$

$$A_p\Omega_p = d_p^2 4\pi \sin^2 \frac{\theta_p}{2}$$
$$\approx d_p^2 H \frac{(INA)^2}{2}$$

$$\approx d_p^2 \frac{H}{8(IFN)^2}$$

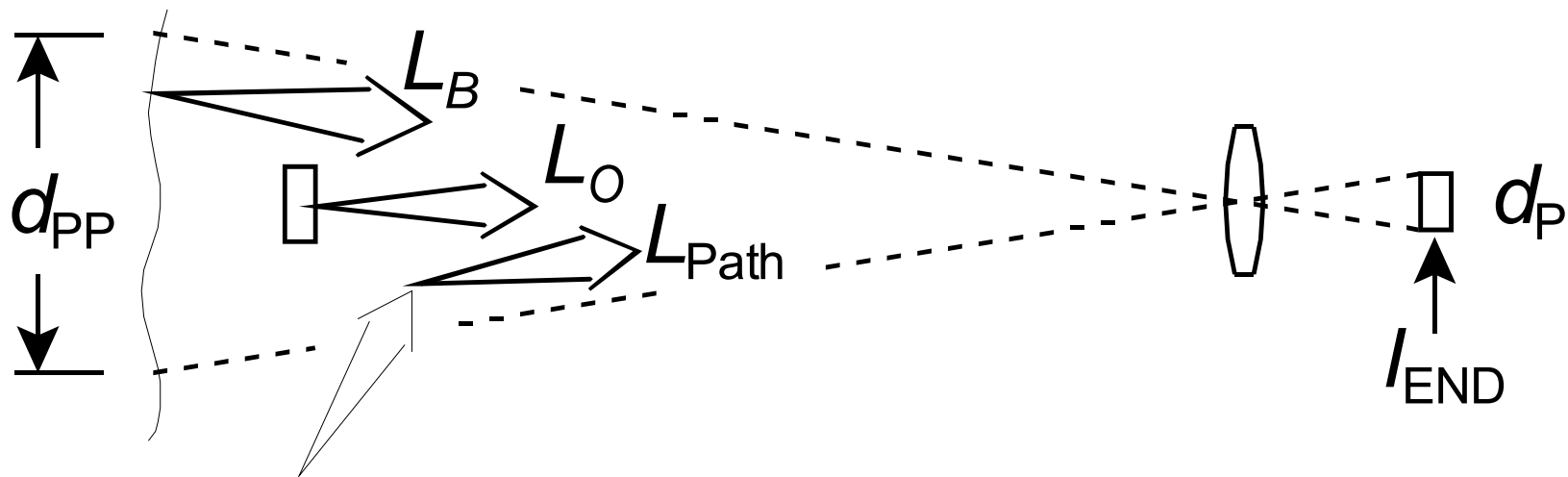
A hemisphere H is reference as

$$H = 2\pi$$



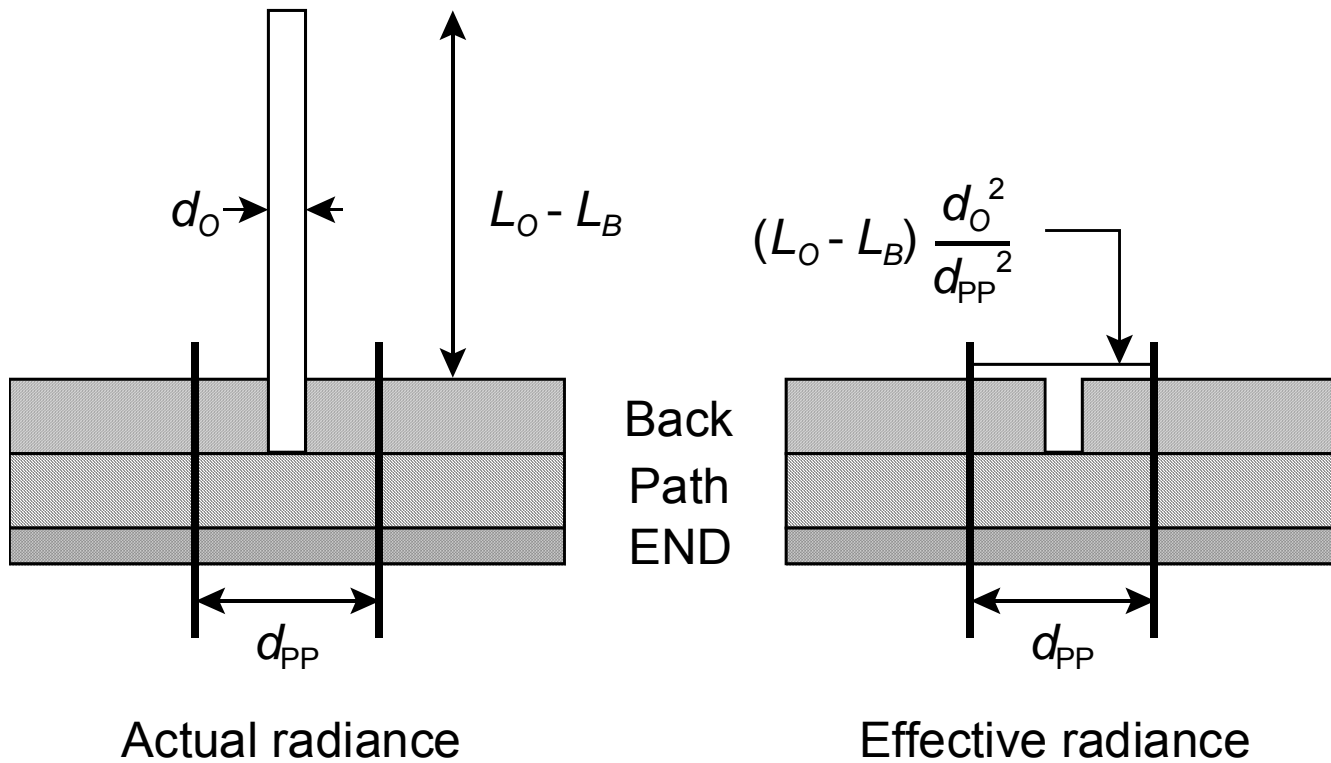
# Path Radiance

- Radiance from path of optics  $L_{Path}$ 
  - Collected from background path of object
  - Collected from foreground path of object
  - Different from background radiance  $L_B$





# Redistribution within pixel



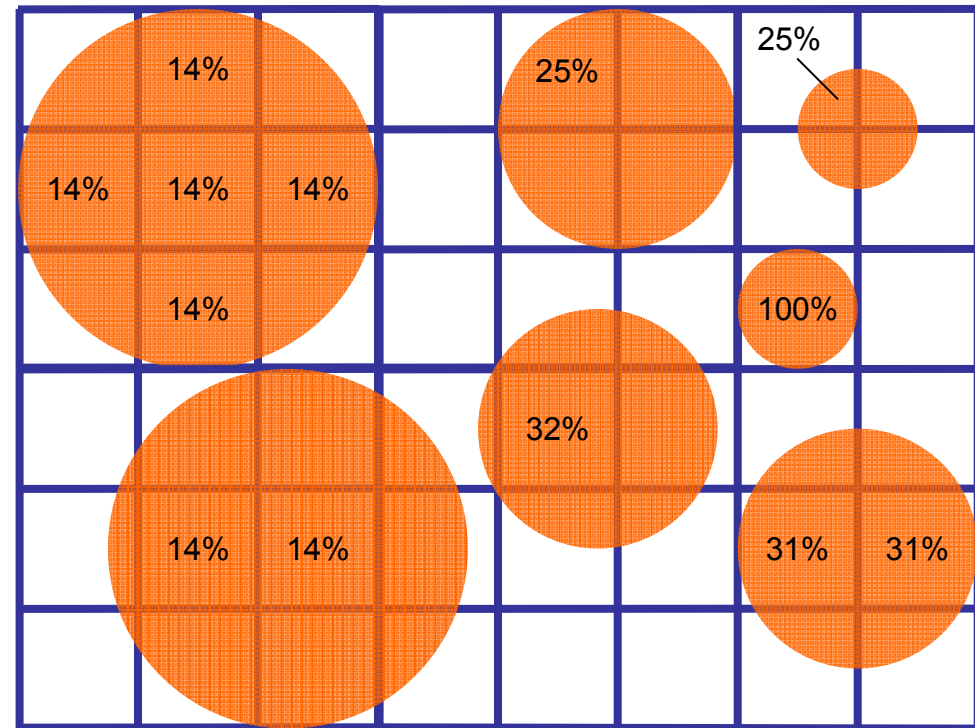
$$C_{\text{SPO}} = \frac{(L_o - L_B) d_o^2 / d_{\text{PP}}^2}{L_B + L_{\text{Path}} + L_{\text{END}}}$$





# Minimum Pixel Collection Efficiency

- Minimum  $\eta_{PC}$ 
  - 14% at 3X pixel-width
  - 25% at 2X pixel-width
  - 25% at 1X pixel-width
- Minimum is at most 25%





## § 6 Aberrations

Aberrations convert signal  
into in background

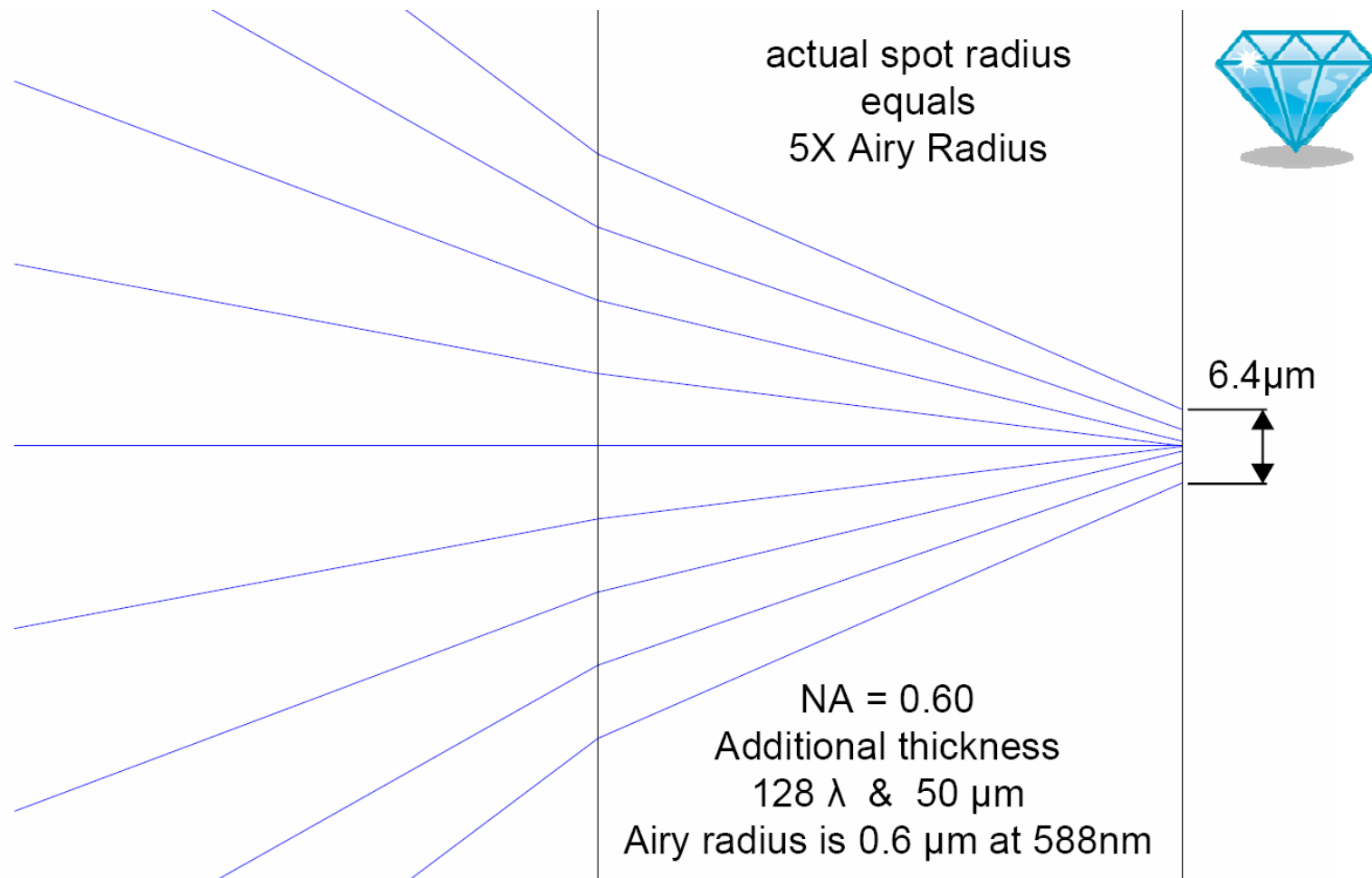


## § 7 Cover aberrations

So much trouble  
created by  
so little thickness

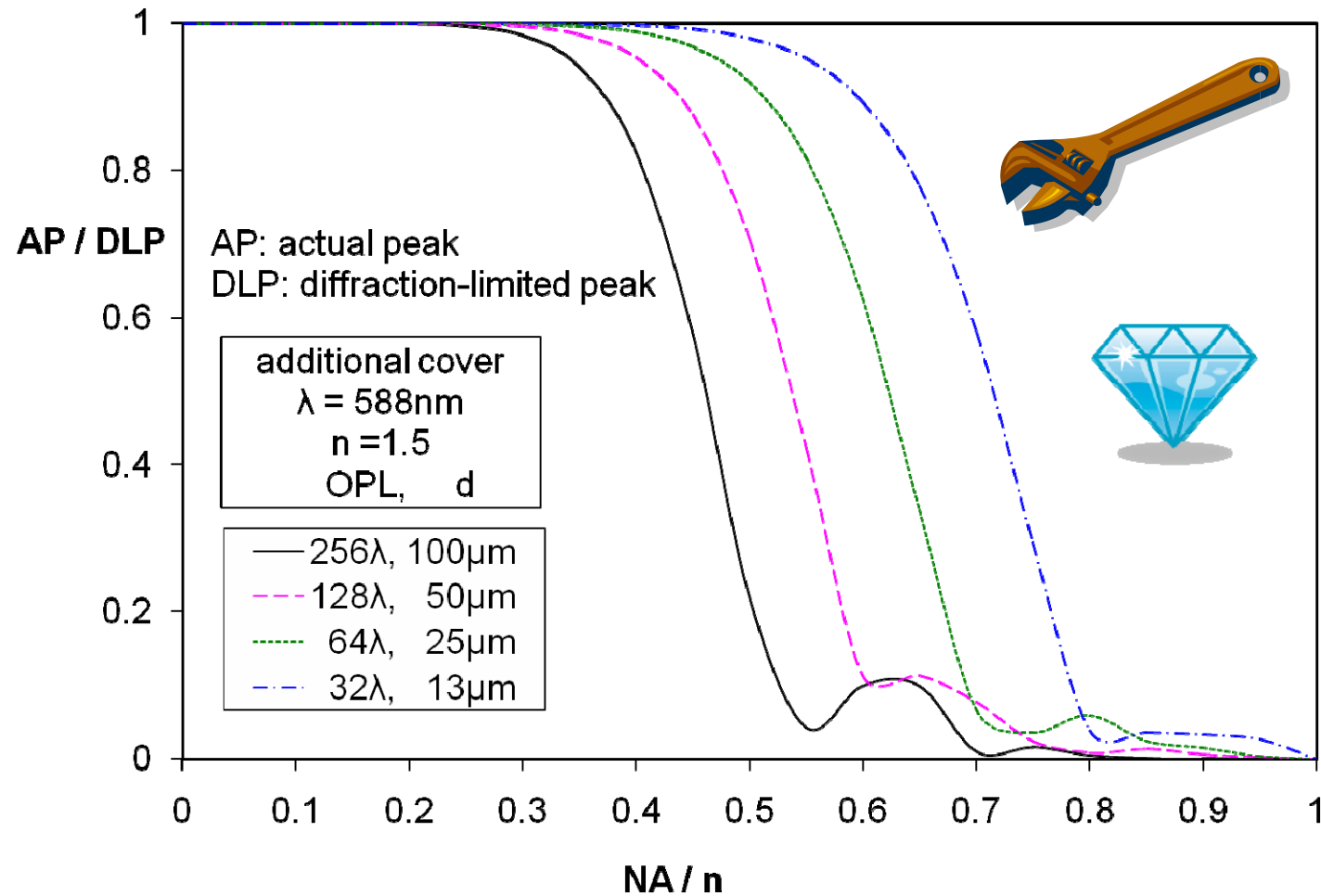


# Cover spherical aberration



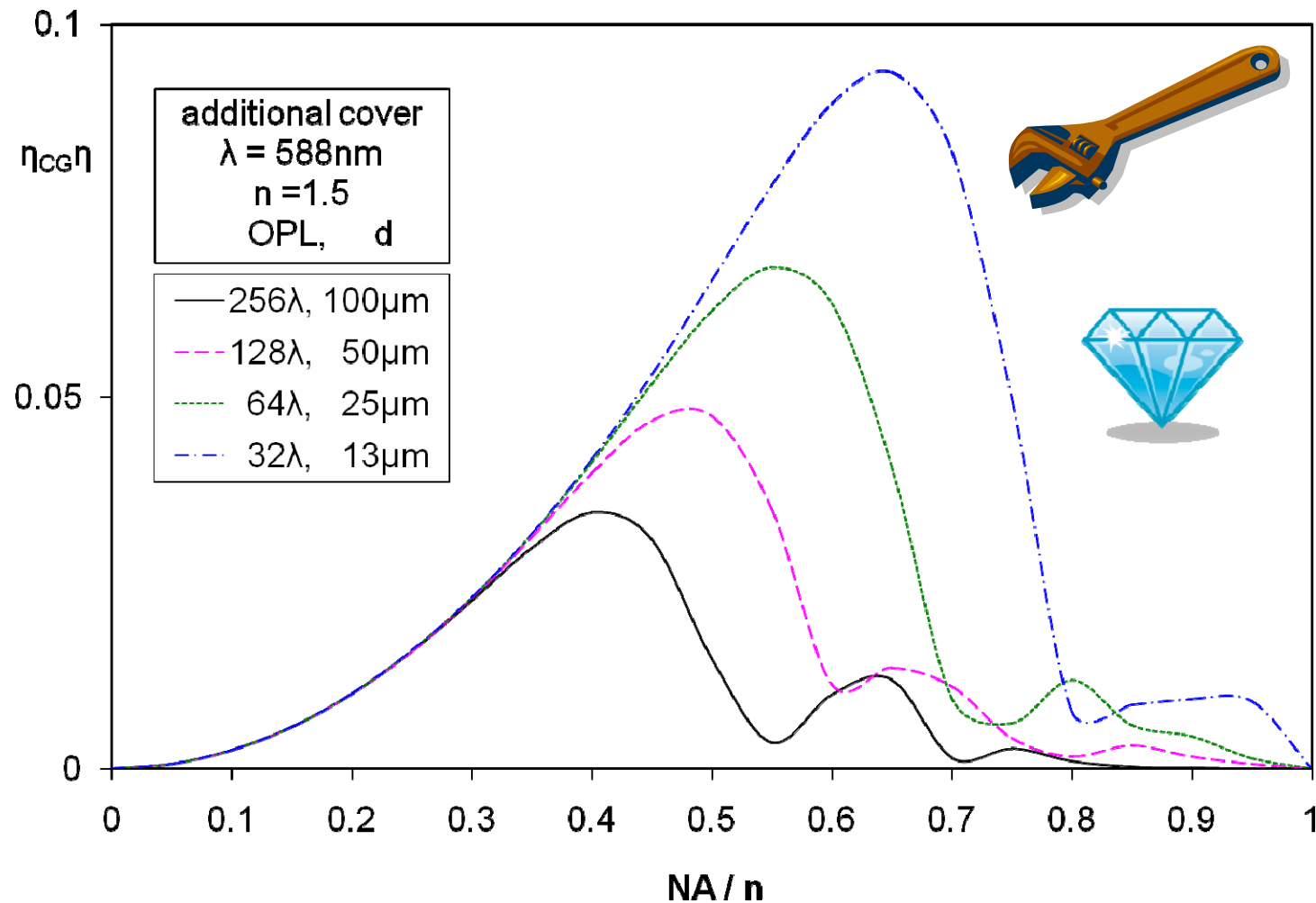


# Cover glass efficiency $\eta_{CG}$





# Combined efficiency $\eta_{CG} \eta_{SC}$





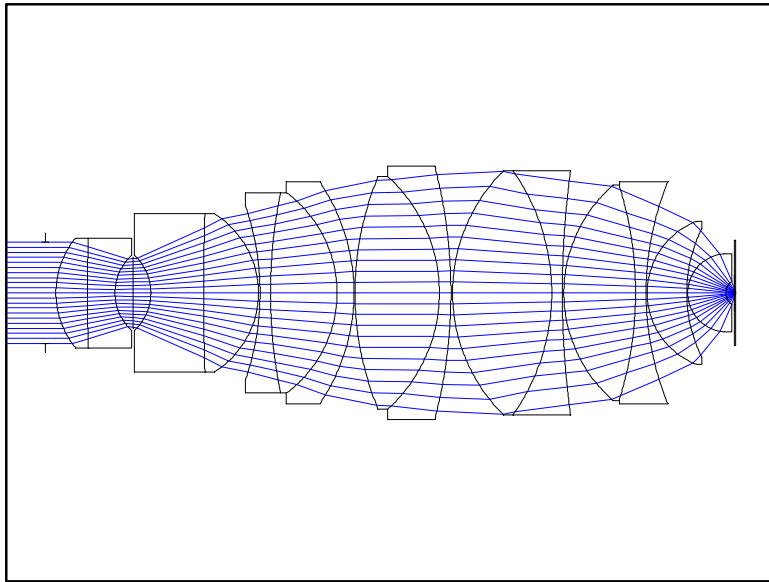
## § 8 Objective aberrations

NA indicates marginal ray  
But not diffraction limit

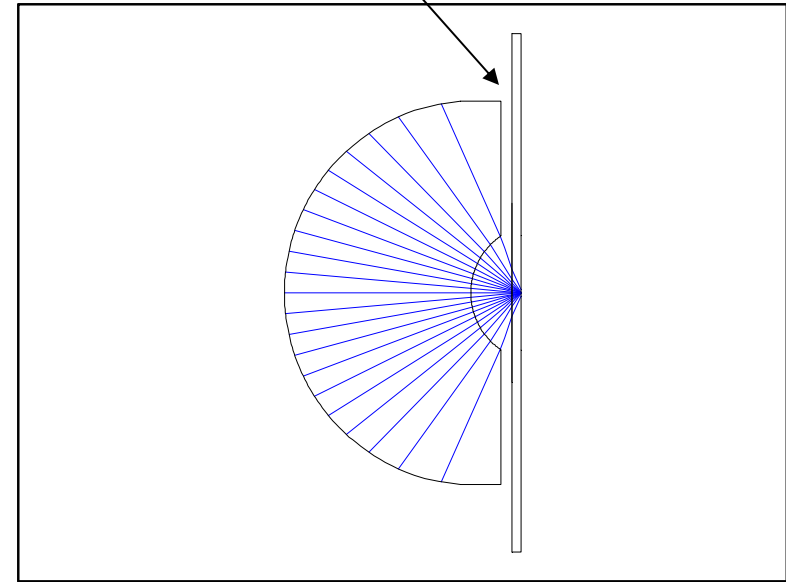


# 60X 1.4NA Immersion Objective

Yamaguchi US pat. 6,519,092 B2



Oil-filled gap

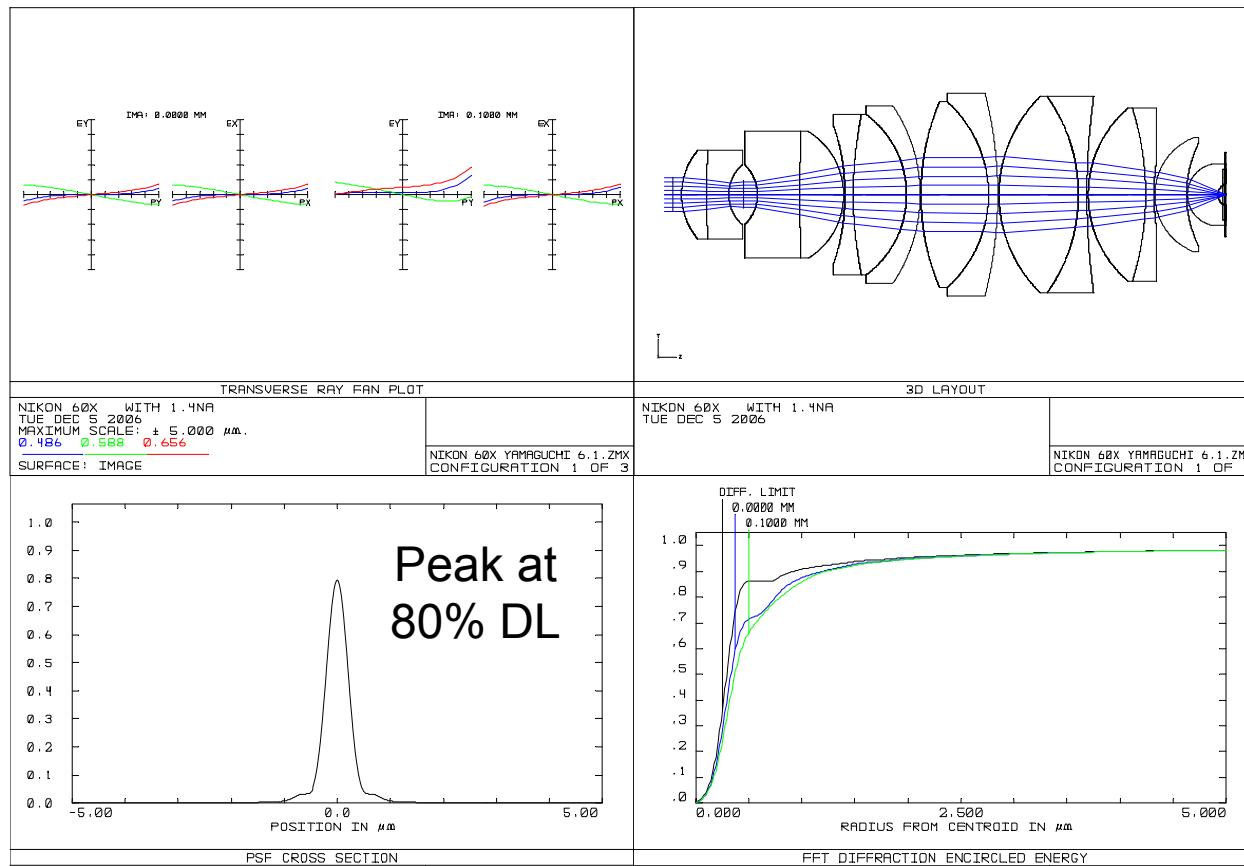


Uniform linear spacing in tube  
nearly corresponds to  
uniform angular spacing at object





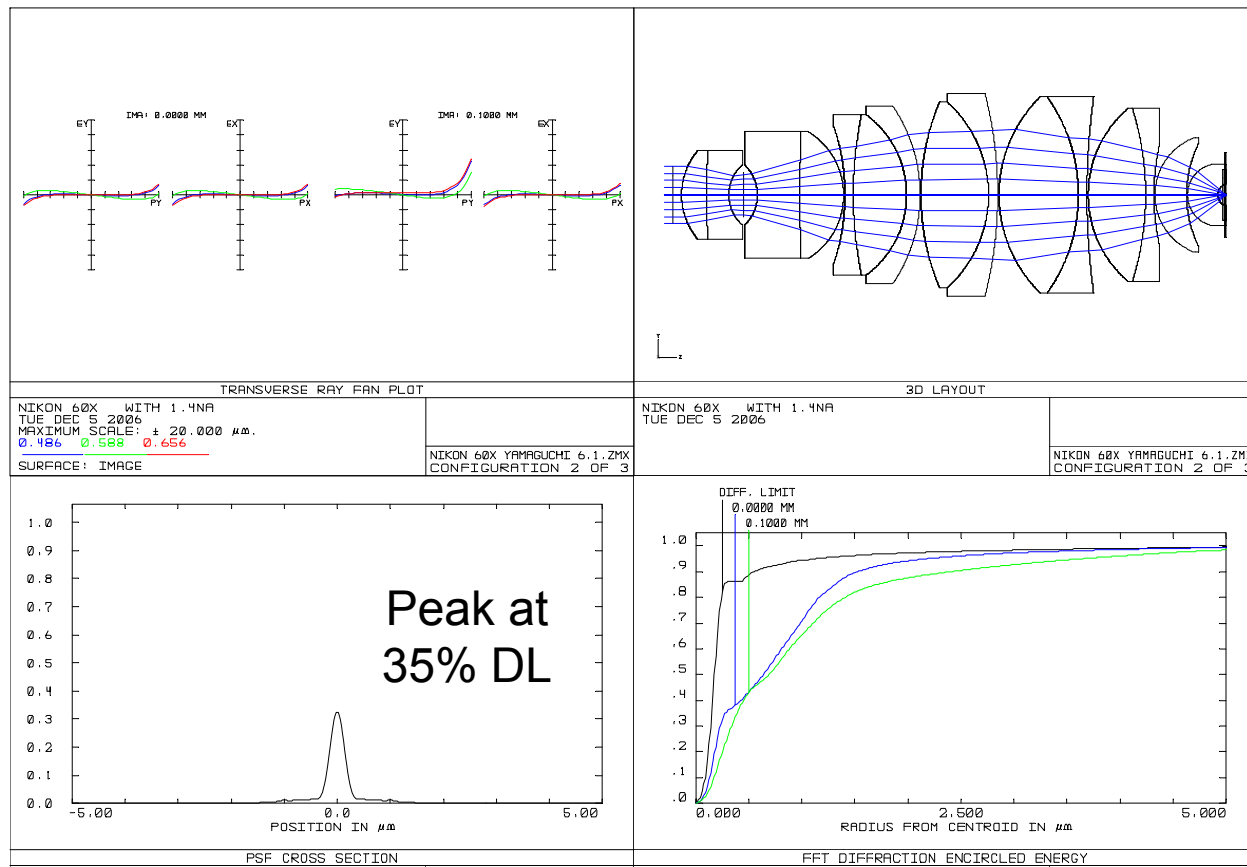
# 60X/1.4 at 0.60NA



Yamaguchi US pat. 6,519,092 B2



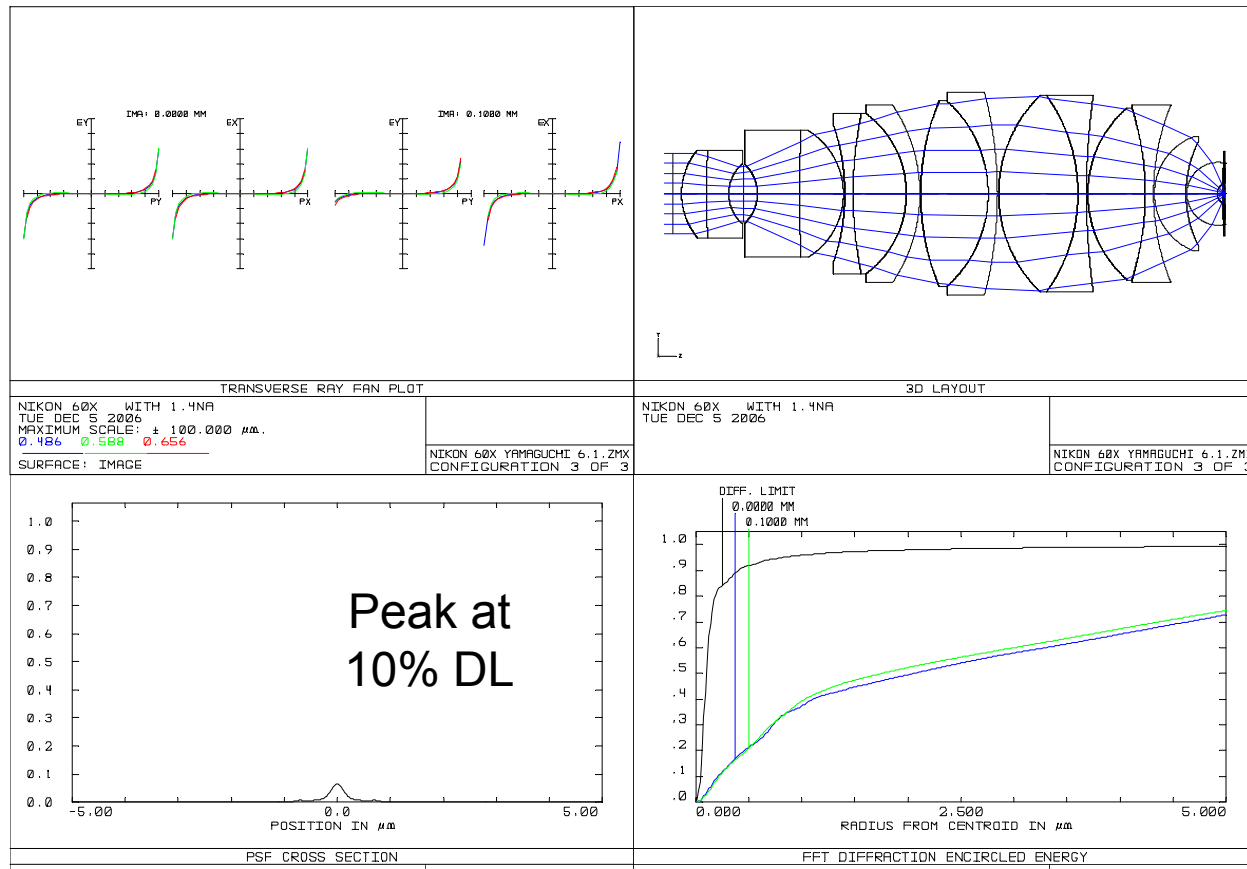
# 60X/1.4 at 1.00NA



Yamaguchi US pat. 6,519,092 B2



# 60X/1.4 at 1.40NA



Yamaguchi US pat. 6,519,092 B2

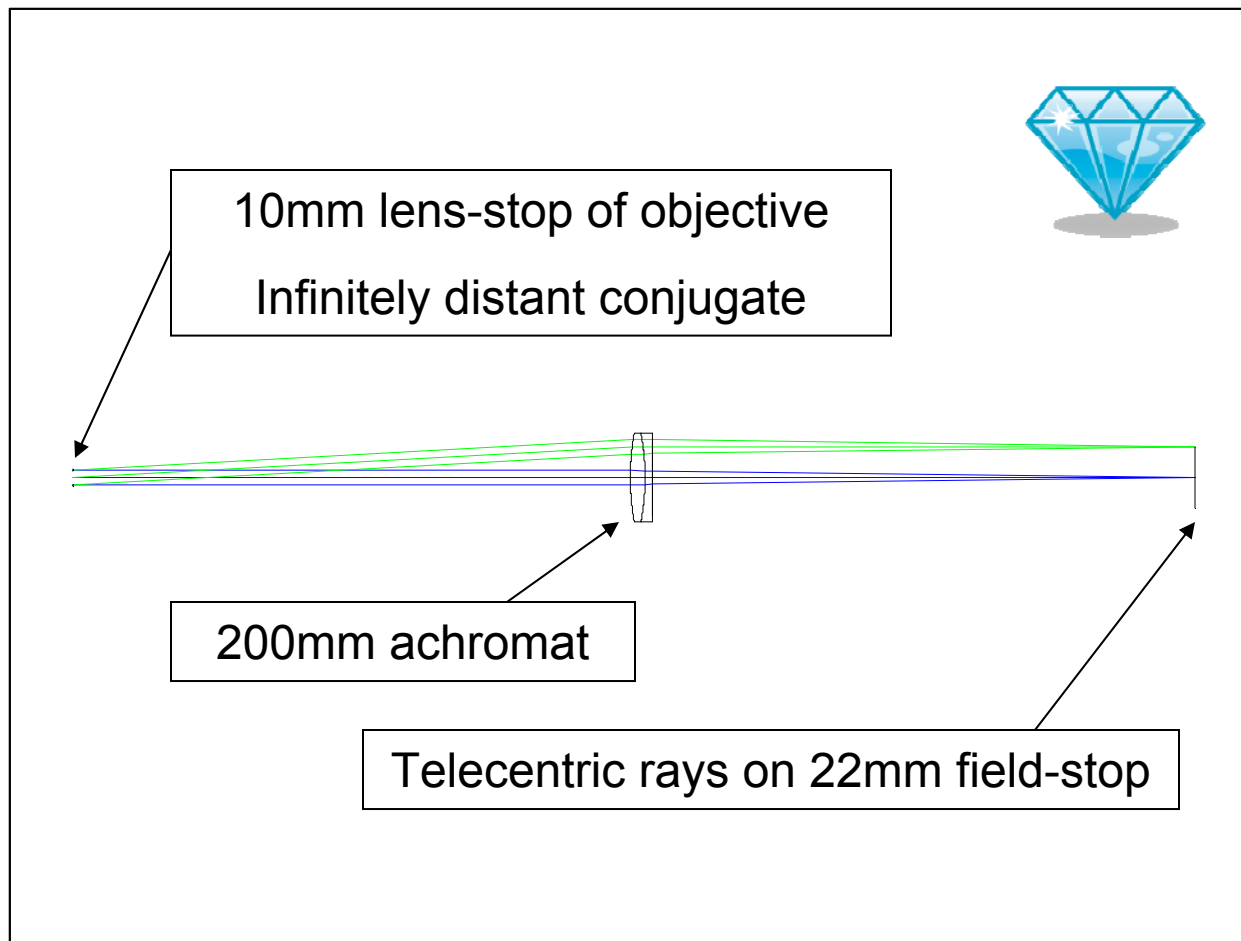


# § 9 Design Example

A custom tube-lens  
for  
an off-the-shelf objective



# Tube lens: Off-the-shelf OTS





# § 10 Conclusion

Closing remarks



# Knowledge is power

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- Comprehension of knowledge
  - Promotes maximum quality
  - Yields lasting value
- Application of knowledge without comprehension
  - Promotes application of sound ideas
  - But not in optimum combination
- Confidence without comprehension
  - Yields trouble!!!



$$C-C = T$$