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Università degli Studi di Trento

Development of a simulation environment for the analysis and the optimal design of fluorescence detectors based on single photon avalanche diodes

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Outlines

- **Introduction**
  - Time-domain fluorescence detection
  - Single photon avalanche diode (SPAD) detectors
- **Model of fluorescence measurement setup**
  - General overview
  - Step by step simulation
- **Validation of the model**
  - Flexibility
  - Prediction ability
- **Conclusions and future work**

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Road Map

Introduction

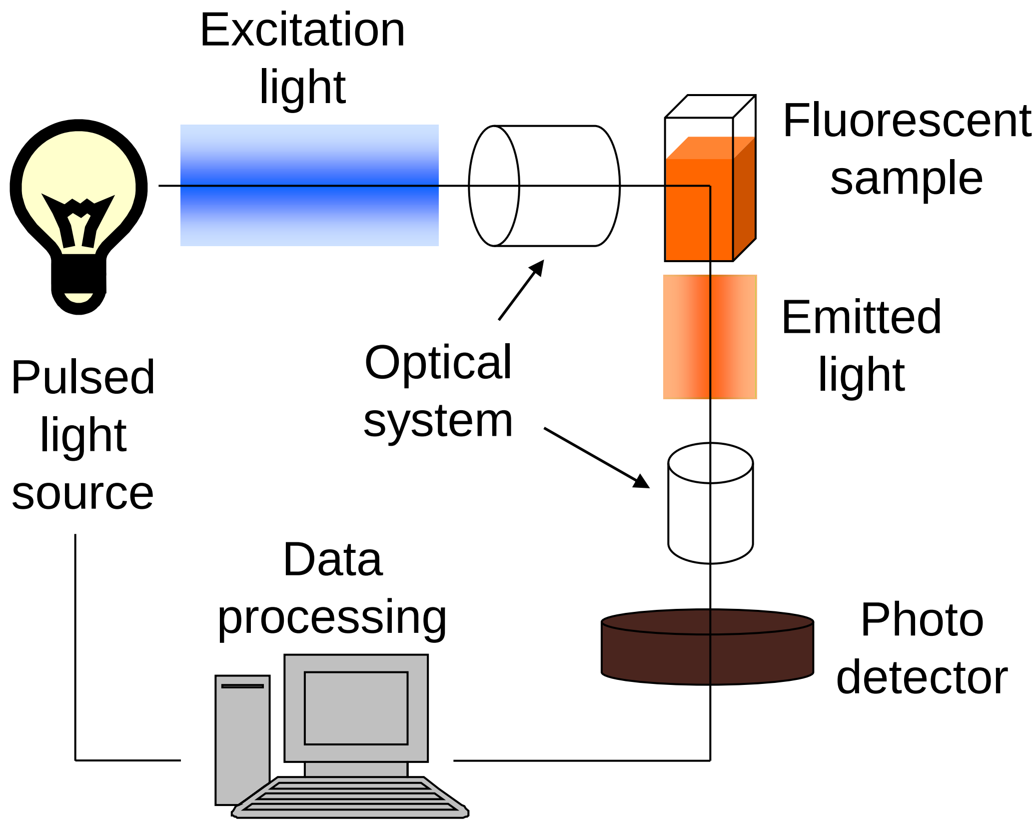
Proposed model

Validation of the model

Conclusions and future work

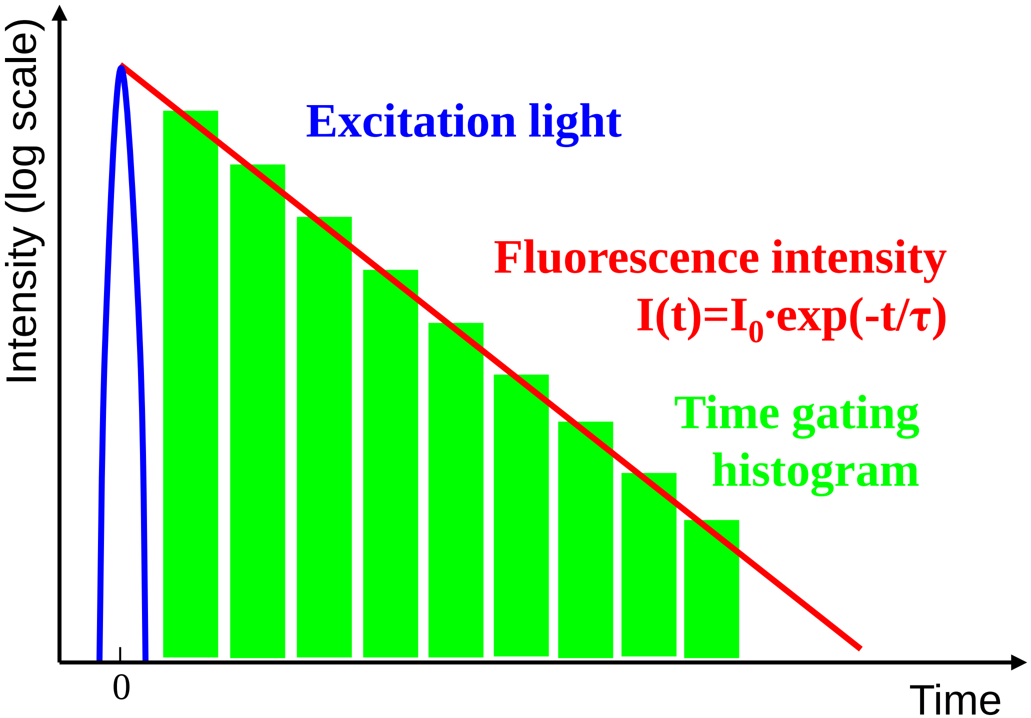
Typical experiment setup

- The photodetector determines the ***accuracy*** **of the measurements.**
- ***General performance*** **of the setup is defined by all part, from light source to lifetime extraction algorithm.**



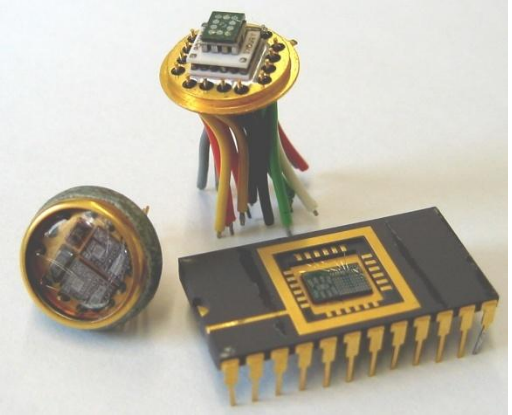
Fluorescence detection

- **Features**
  - Nanosecond time scale
  - Typical lifetime 1ns – 20ns
  - Single photon sensitivity
- **Time-correlated single photon counting (TCSPC)**
  - Pulsed light source
  - Time-stamp of the first detected photon
- **Time-gating**
  - From 2 time intervals
  - More than one photon per pulse



Single photon avalanche diode

SPAD is a class of solid-state photodetectors based on a reverse biased p-n junction.



Advantages

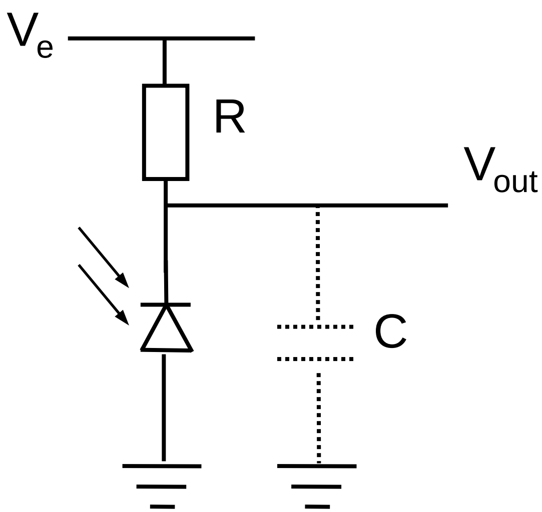
- Small size
- Low cost
- Magnetic field immunity
- Low voltage supply
- Suitable for matrices
- Real time data processing (only for CMOS SPAD)

Disadvantages

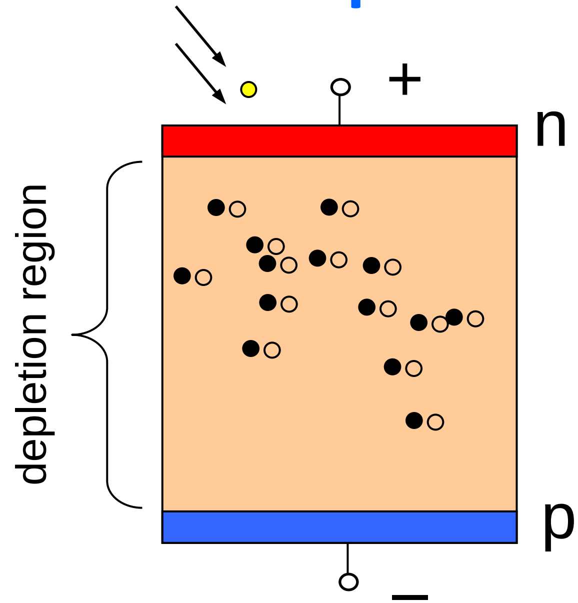
- Higher noise
- Narrow absorption spectrum

Photon detection

Simplified SPAD diagram



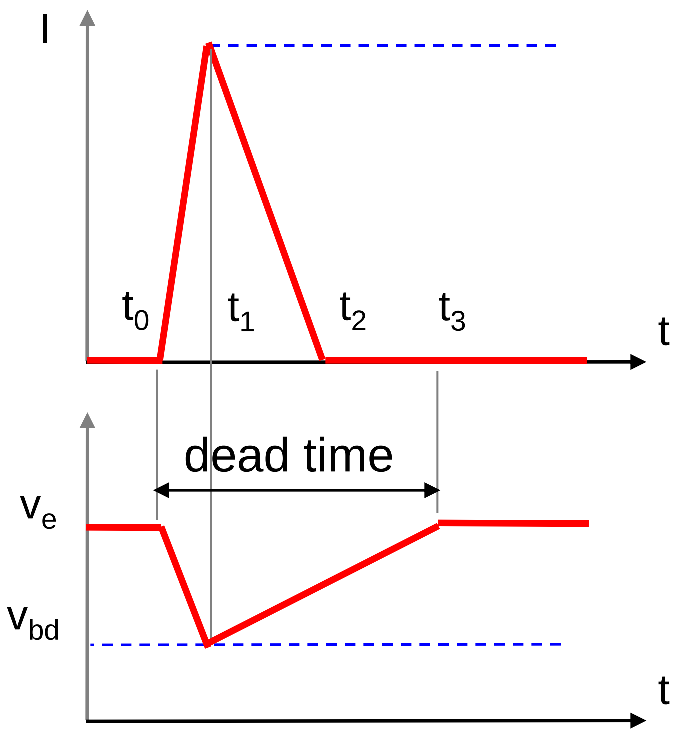
Avalanche process



- Excess bias voltage V_e is applied to SPAD
- Electric field in depletion region is very high
- Single photon can create an avalanche

Timing

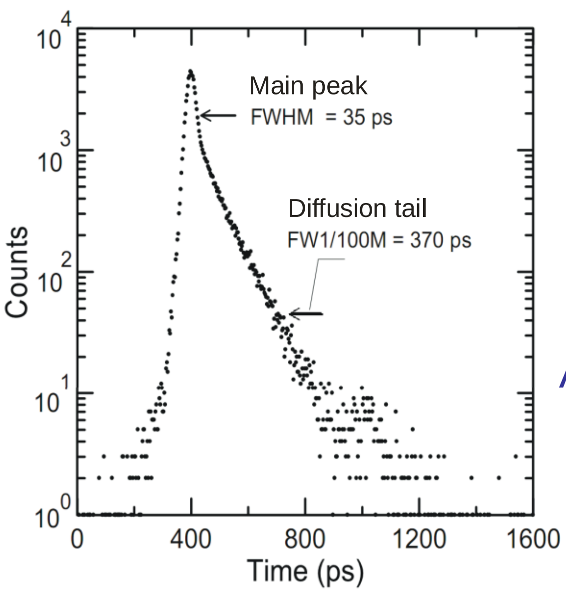
Avalanche current and voltage



Timing

- t1-t0 – time jitter
- t2-t1 – quenching time
- t3-t1 – recharging time

Typical time jitter curve



A. Gulinatti et al., Proc. ESSDERC (2005)

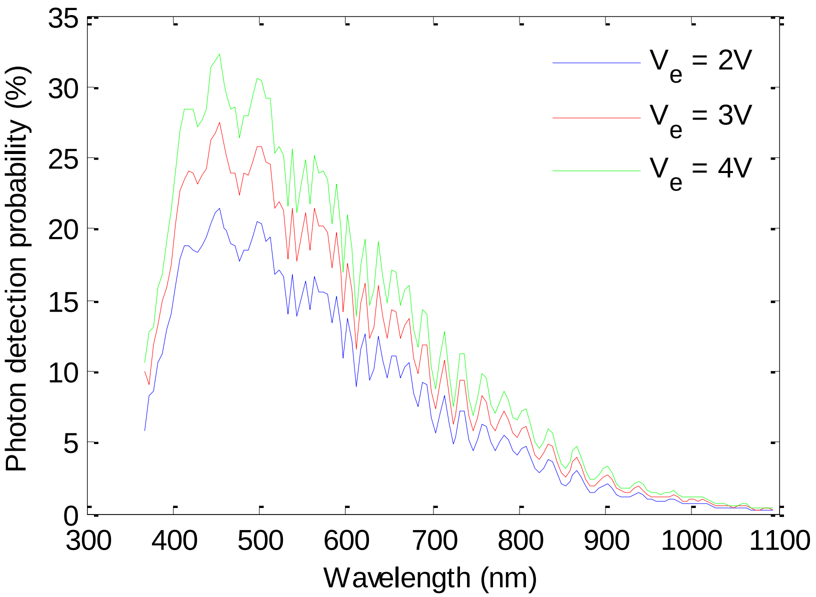
Photon detection probability

$$PDP = \frac{\text{output current pulses}}{\text{incoming photons}}$$

$$PDP = QE(\lambda)*P_{av}(V_e)$$

QE – quantum efficiency

P_{av} – avalanche triggering probability by the primary e-h pair

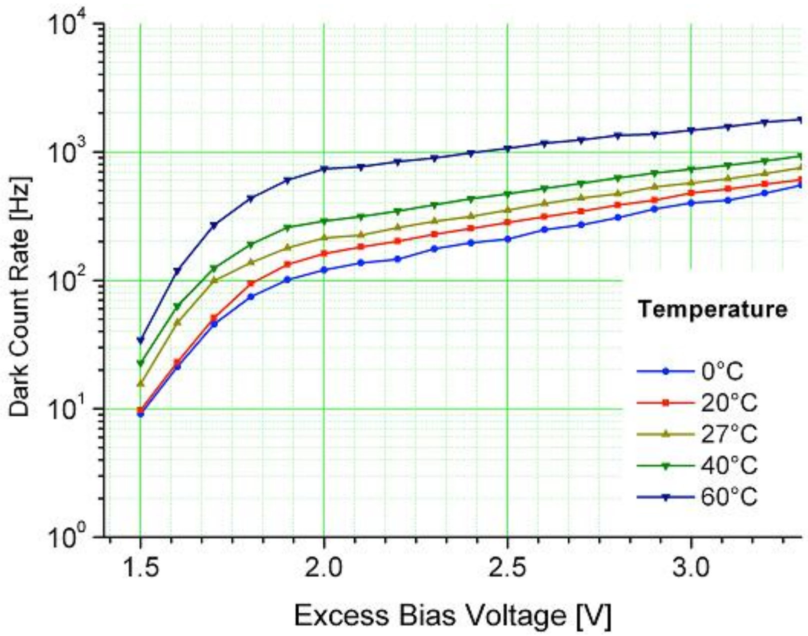


D. Stoppa et al., IEEE Sens. J. (2009)

Detector noise

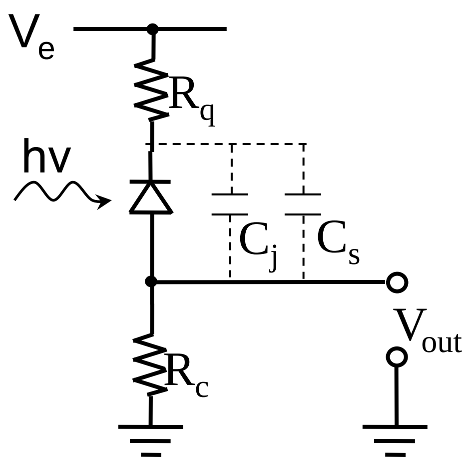
Dark count rate (DCR) – avalanche triggering rate of the detector held in the darkness

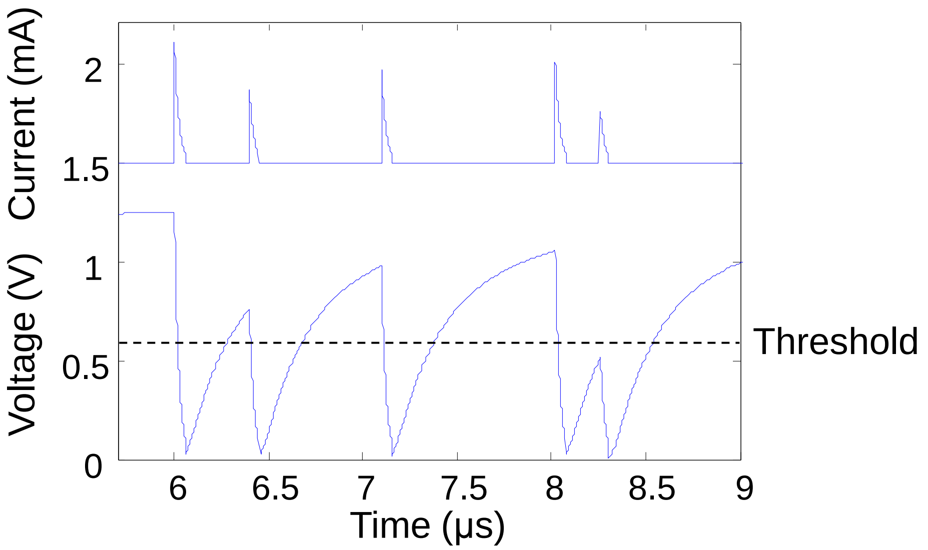
- – Thermal generation
- – Band to band tunneling
- – Afterpulsing



C. Niclass et al., SPIE Optics East (2006)

Passive quenching

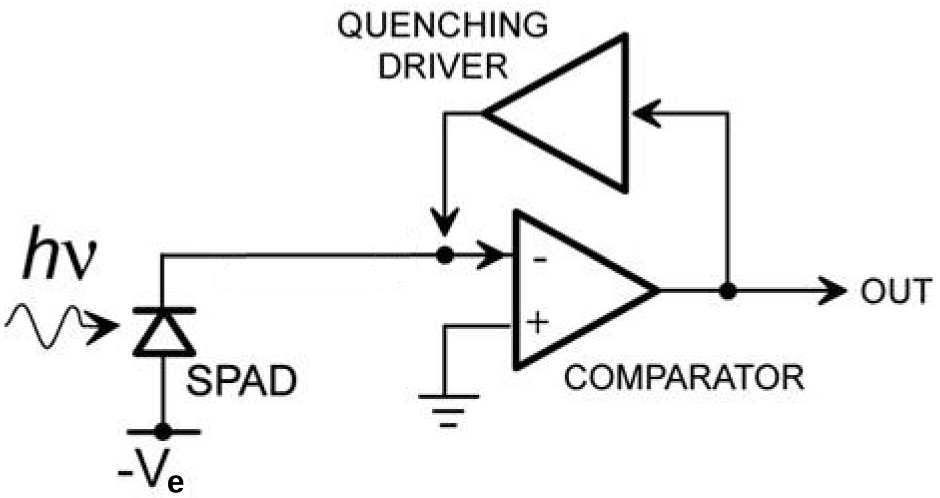




- **Simple to organize**
- **Long recovery time**
- **Triggering probability depends on time**
- **Not constant dead time**
- **Low maximum count rate**

Active quenching

- Short recovery time
- Fixed and controlled dead time
- Adjustable hold-off time and afterpulsing
- High maximum count rate



S. Cova et al., J. Mod. Opt. (2004)

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SPAD in easy terms (video)

SPAD performance

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Characteristic	0.18µm CMOS	InGaAs/InP	Double-epitaxial	130nm CMOS
Area	2µm x2µm	Ø 25µm	Ø 50, Ø 200 µm	Ø 8µm
Dead time	3ns			
DCR		70 kHz	1kHz, 50kHz	20Hz
PDP		45% at 1310nm	55% at 500nm, 68% at 550nm	20%-25% at 440-570nm
Time response	26.7ps (96.1ps)	30ps	35ps	200ps

Characteristic	HV CMOS	0.35µm HV CMOS	130nm CMOS	0.35µm CMOS
Area	Ø 5µm	15.8 x 15.8µm	Ø 10µm	Ø 10µm
Dead time		200ns		100ns
DCR	50Hz	1kHz	100kHz	600Hz
PDP	34% at 470nm	32% at 450nm	34% at 450nm	35% at 460nm
Time response	80ps	160ps	144ps	

Motivation

- Different manufacturing techniques result in SPADs with different characteristics
- Current works focus on particular parameters out of system context
- The importance of particular detector characteristic depends on application
- Trial-and-error is expensive and time consuming

How can we estimate the suitability of a SPAD-based detector for specific application?
(without producing it)

Goal

Create a tool that will allow the performance analysis and optimization of a SPAD-based system

– Considering all essential parts of fluorescence measurement experiment

– Adjustable to different experiments

Expected benefits

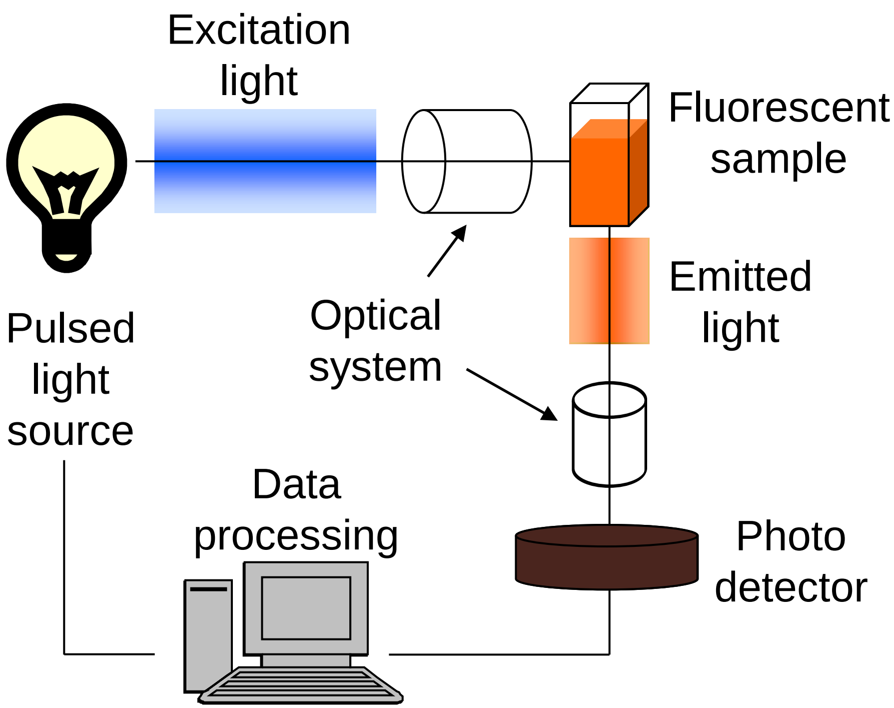
The tool will enable the researchers to…

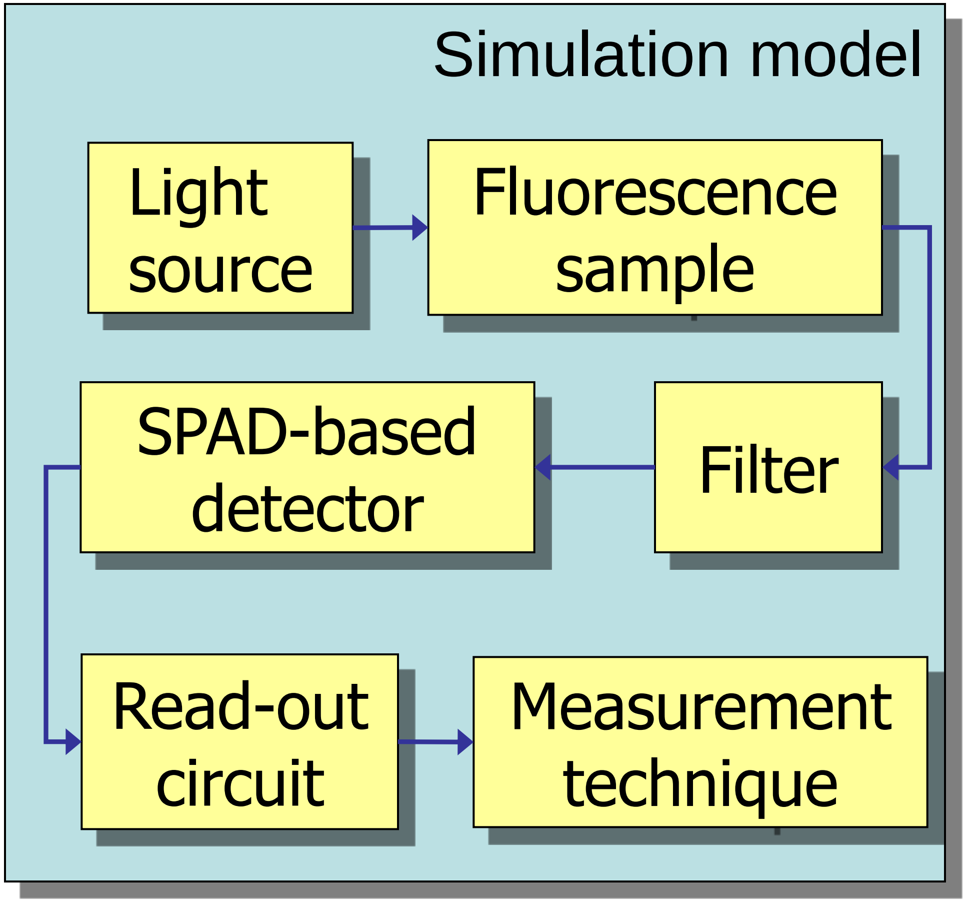
- Predict the results of planned experiments
- Explain the results of real experiments
- Determine the optimal trade-off between opposing characteristics
- Generate test data for evaluation of novel data processing algorithms

Road Map

- Introduction
- **Proposed model**
- Validation of the model
- Conclusions and future work

Proposed model





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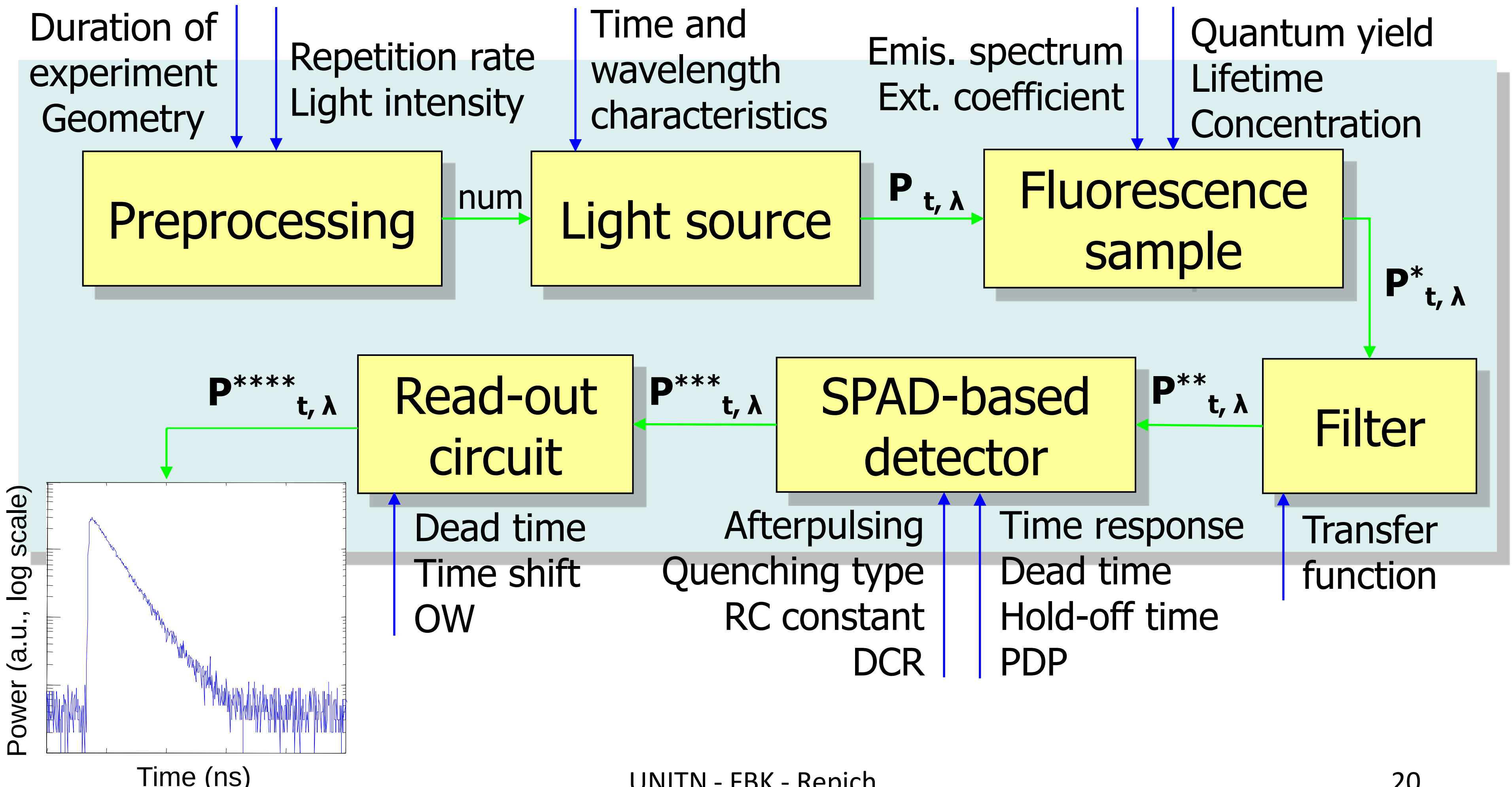
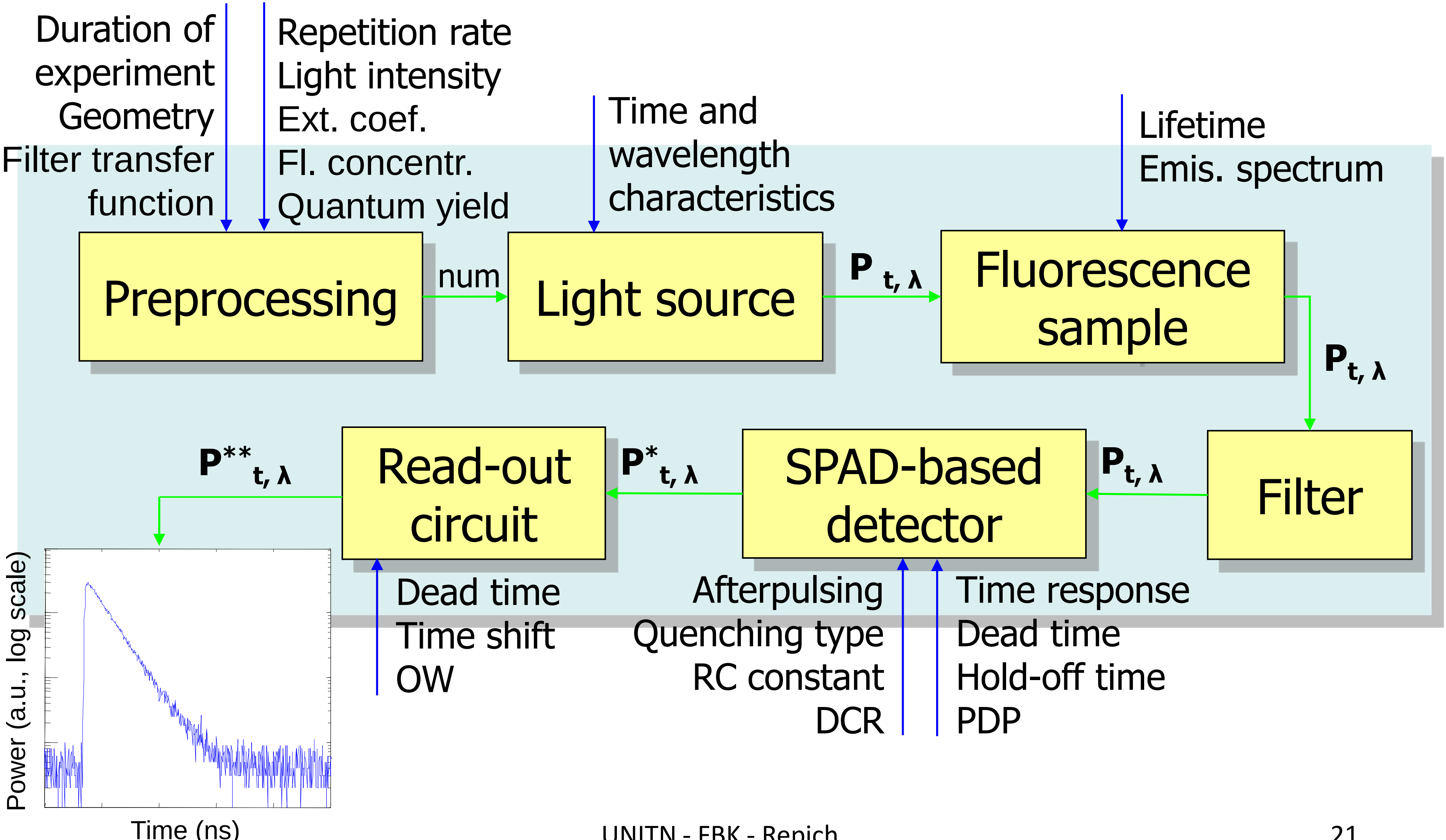
Simulation workflow (forward type)

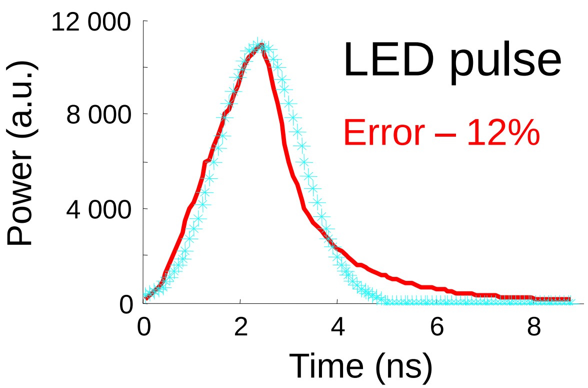


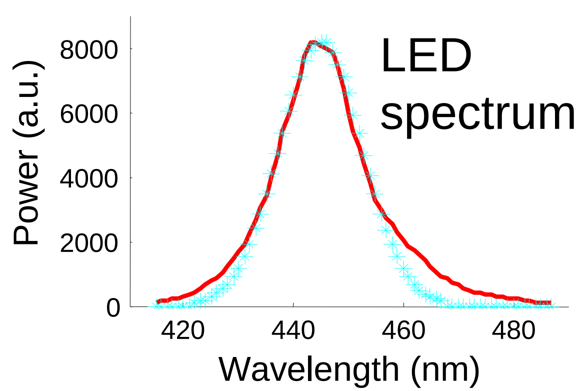
Simulation workflow (backward type)

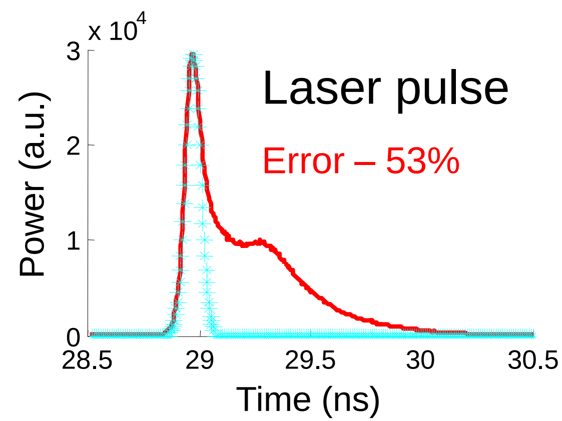


Light source simulation

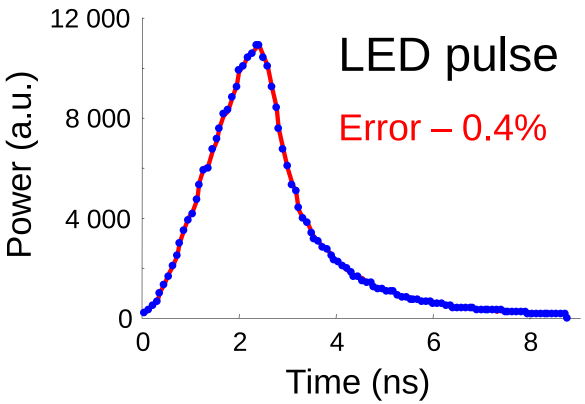
- **Gaussian approximation**

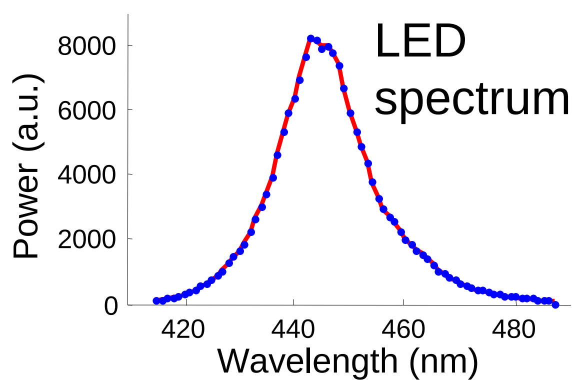


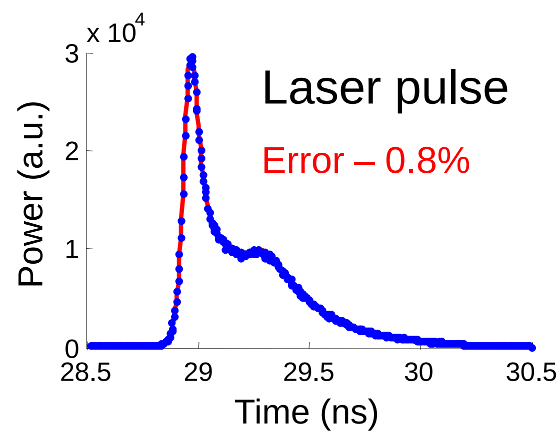




- **Tabulated functions**





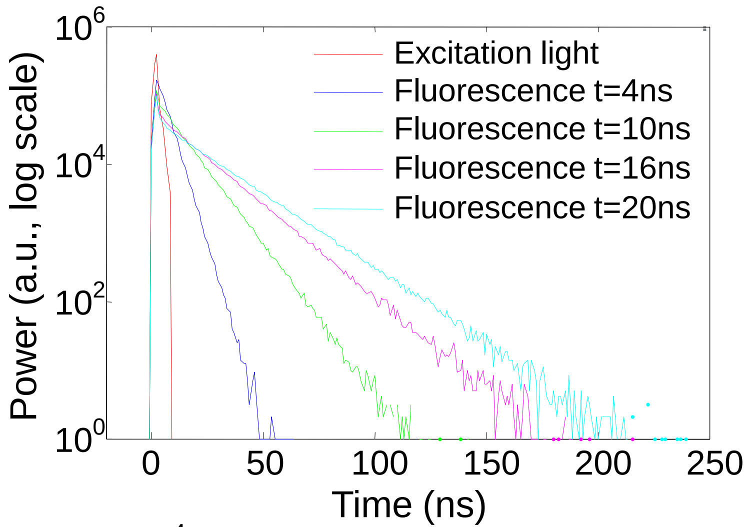


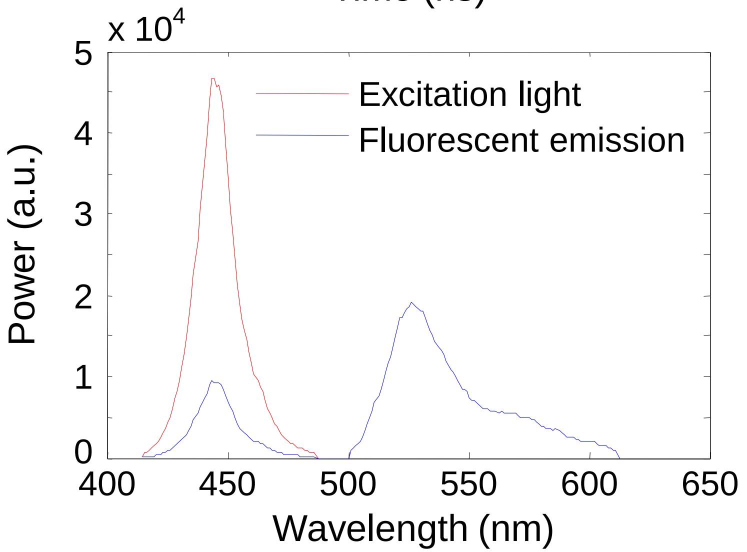
Measured Simulated

Fluorescence simulation

Assumptions:

- Beer-Lambert law
- Uniform fluorophore distribution
- Negligible optical density
- Monoexponential decay
- No other processes
- Light source as a point





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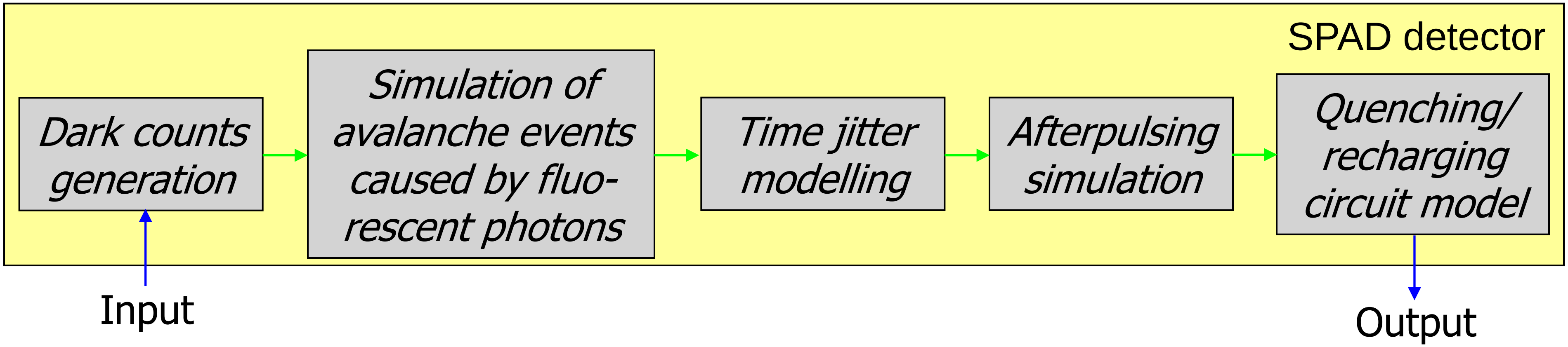
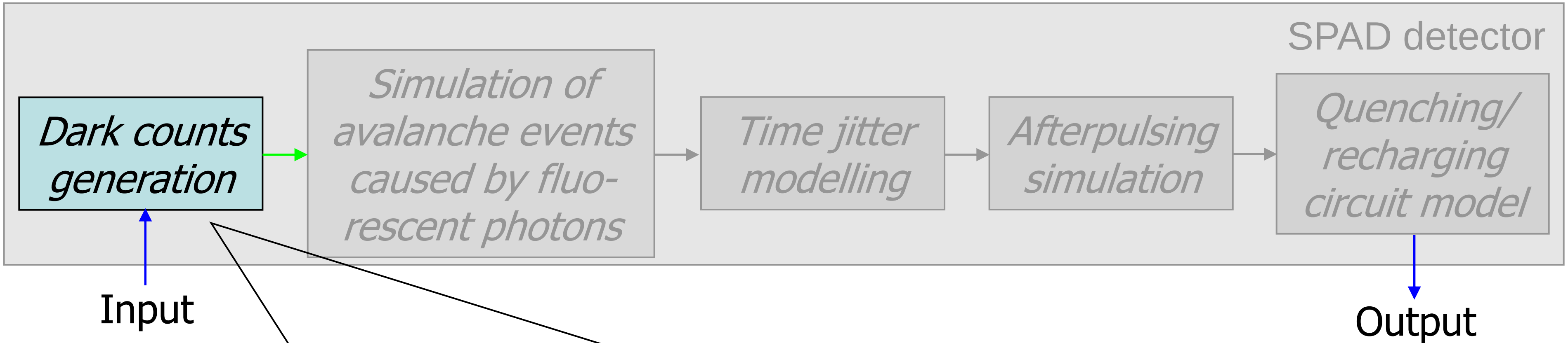
SPAD simulation



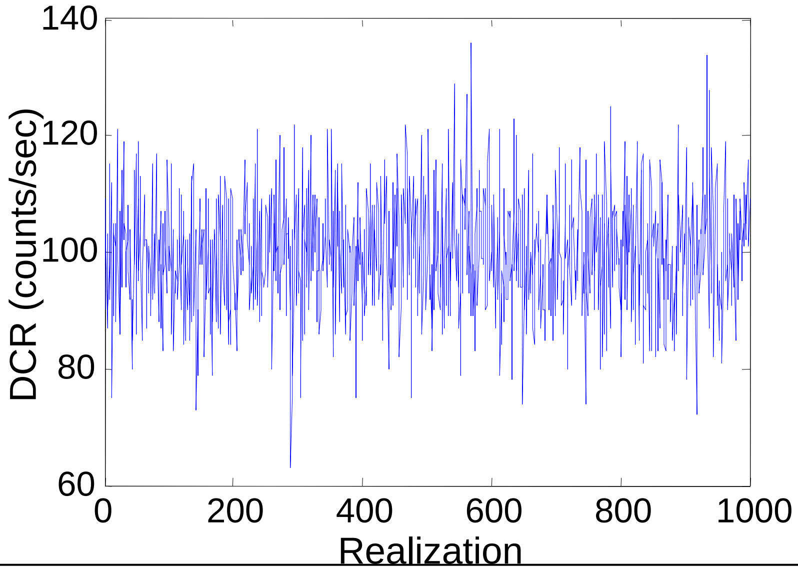
Internal noise



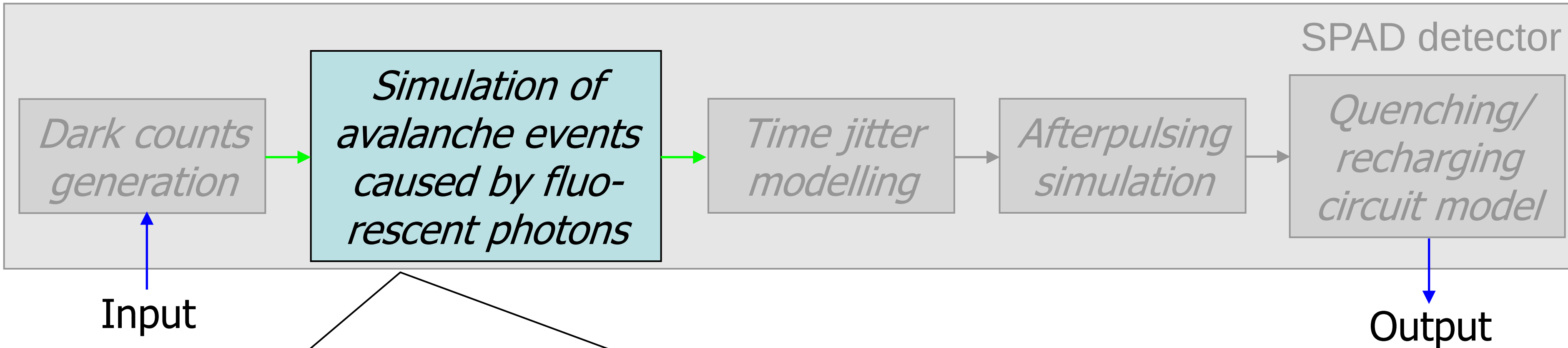
- **Poisson flow**

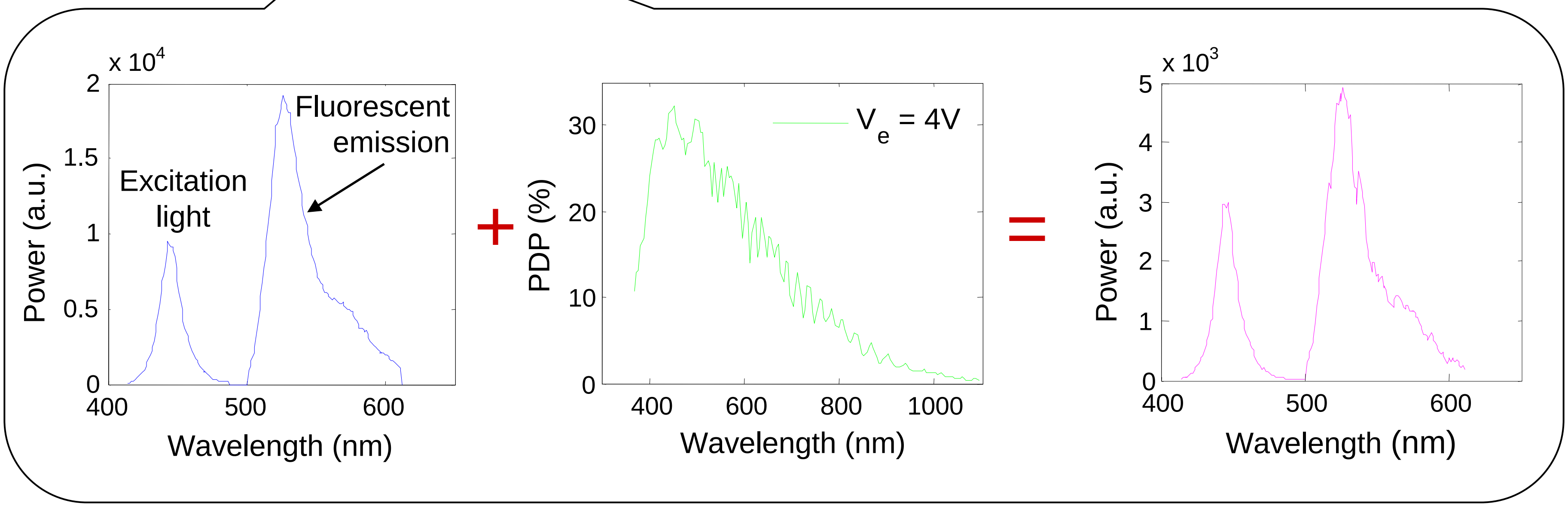
$$P(\mu = n, t) = \frac{\lambda^n}{n!}\exp(-\lambda t)$$

where λ = DCR

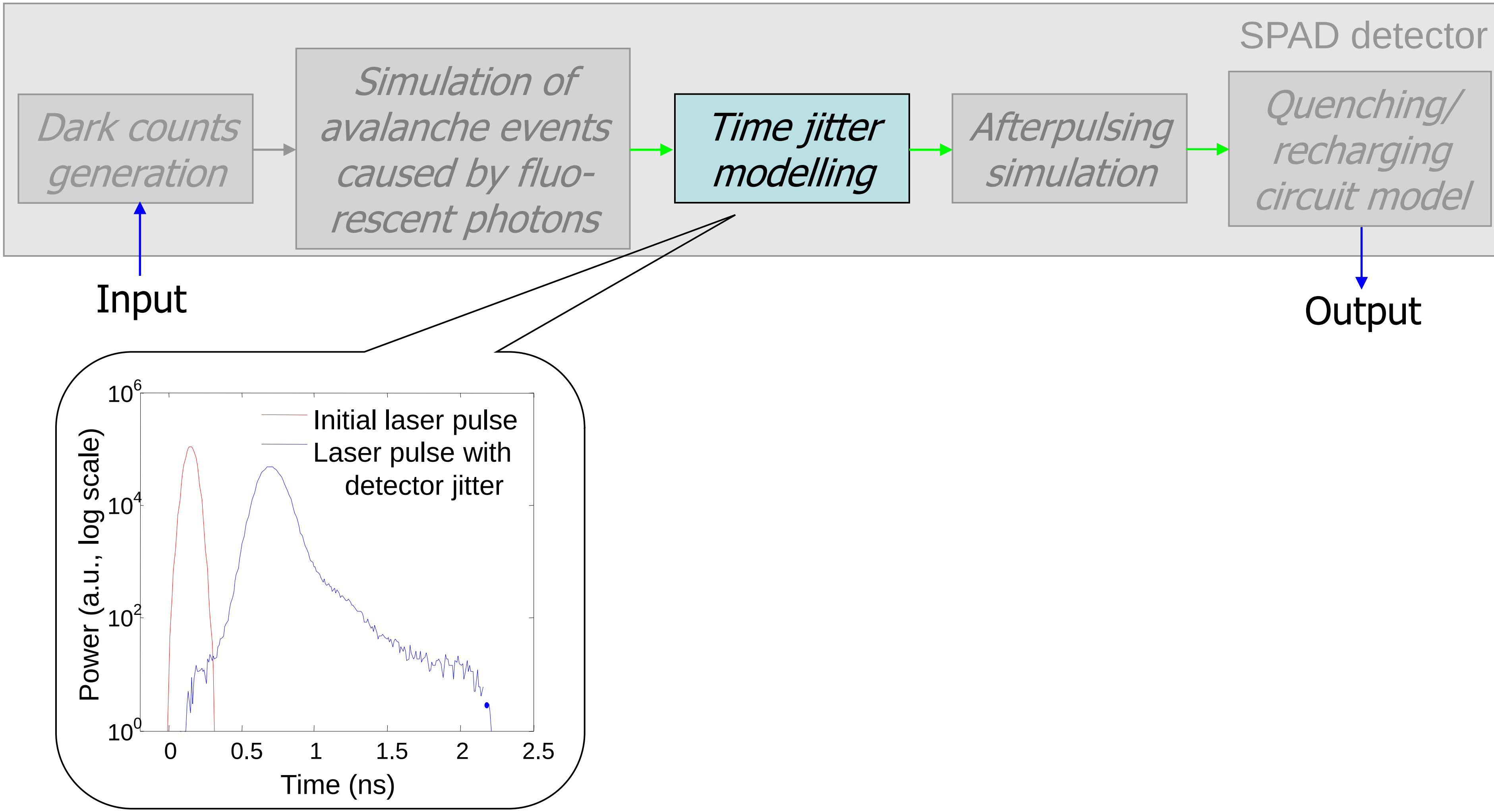


Photon detection probability

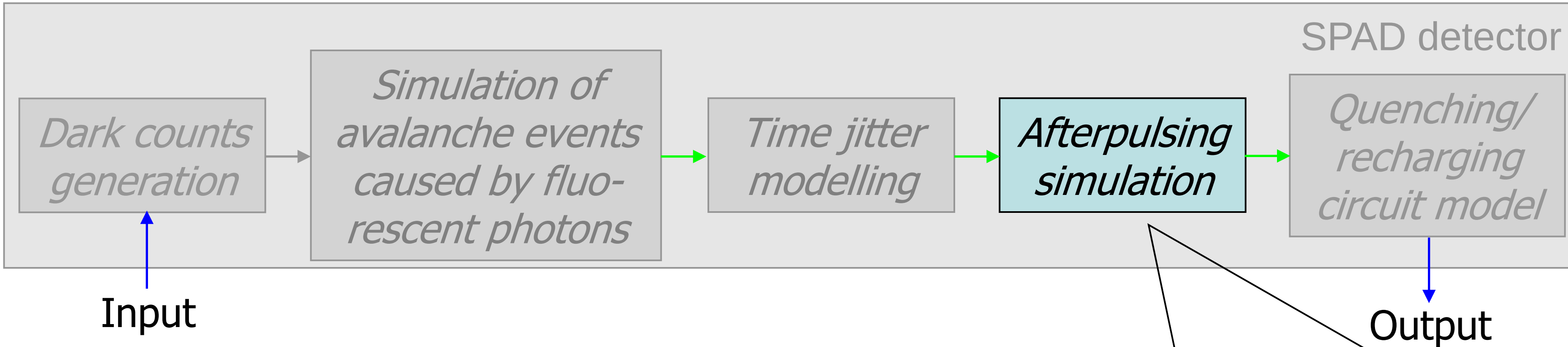


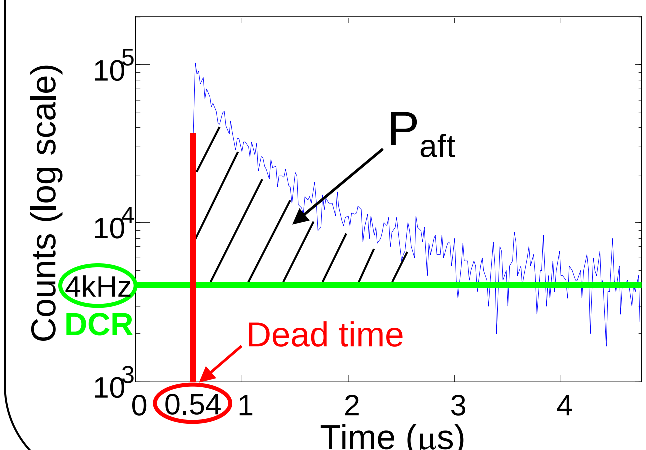


Time jitter


SPAD detector
Dark counts generation
Simulation of avalanche events caused by fluo-rescent photons
Time jitter modelling
Afterpulsing simulation
Quenching/ recharging circuit model
Input
Output
Initial laser pulse
Laser pulse with detector jitter
Power (a.u., log scale)
Time (ns)
10^6
10^4
10^2
10^0
0
0.5
1
1.5
2
2.5

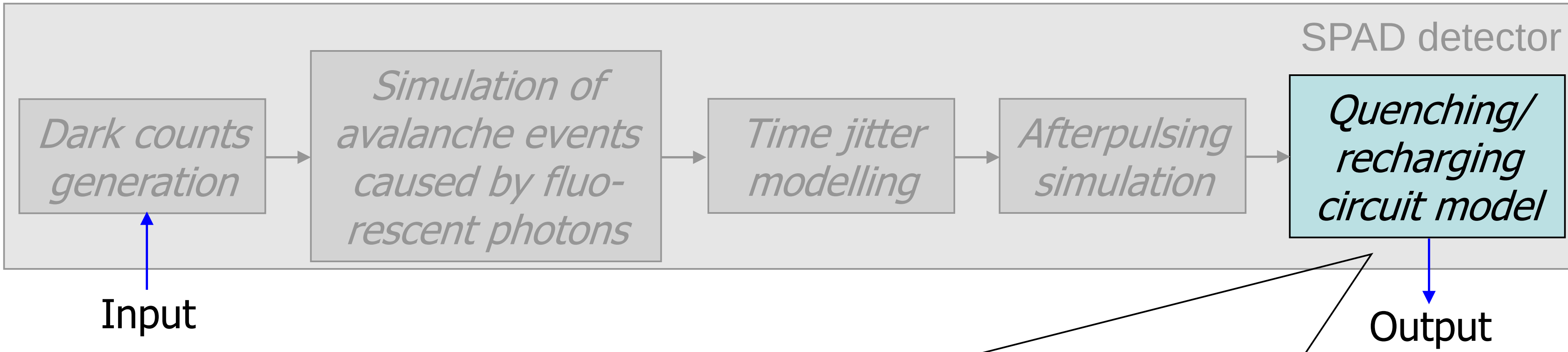
Afterpulsing





- **by rejection method**
- **for each fluorescence photon**
- **according to afterpulsing curve**

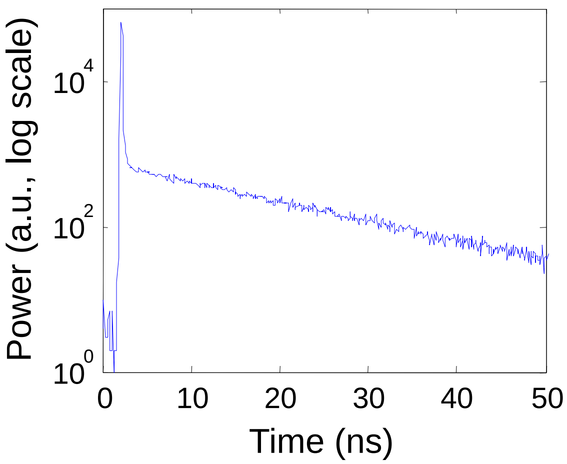
Quenching/recharging circuit

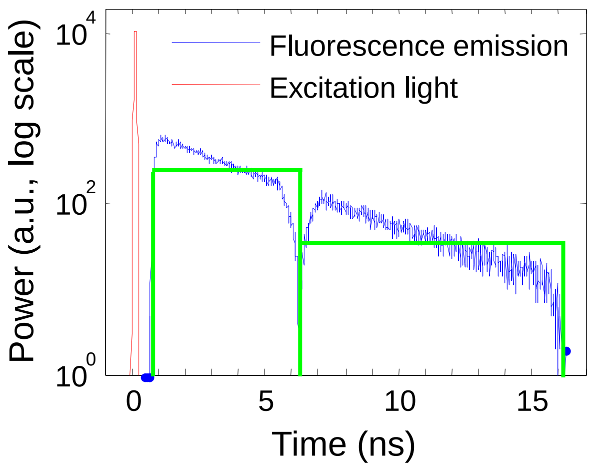


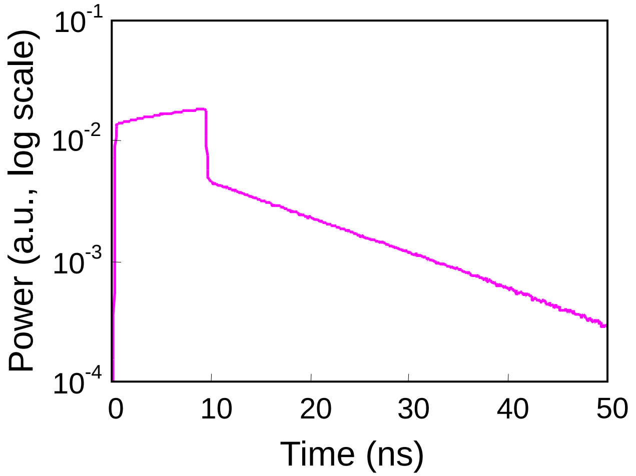
- **Active quenching**
  - if $E_{i+1} < E_i$ + dead time –> delete $(i+1)^{th}$ event
  - E can be fluorescent photon detection, dark photon generation or afterpulse
- **Passive quenching**
  - Δt between consecutive events
  - $V_i = V_{\max} \exp(-RC/\Delta t_i)$
  - $V_i < V_{th}$ –> delete i^{th} event

Measurement technique simulation

- **TCSPC**
  - first photons
  - several photons
- **Time-gating**
  - all photon in dedicated time intervals
- **Modified time-gating**
  - all photons in one observation window (OW)
  - shift OW







Road Map

Introduction

Proposed model

Validation of the model

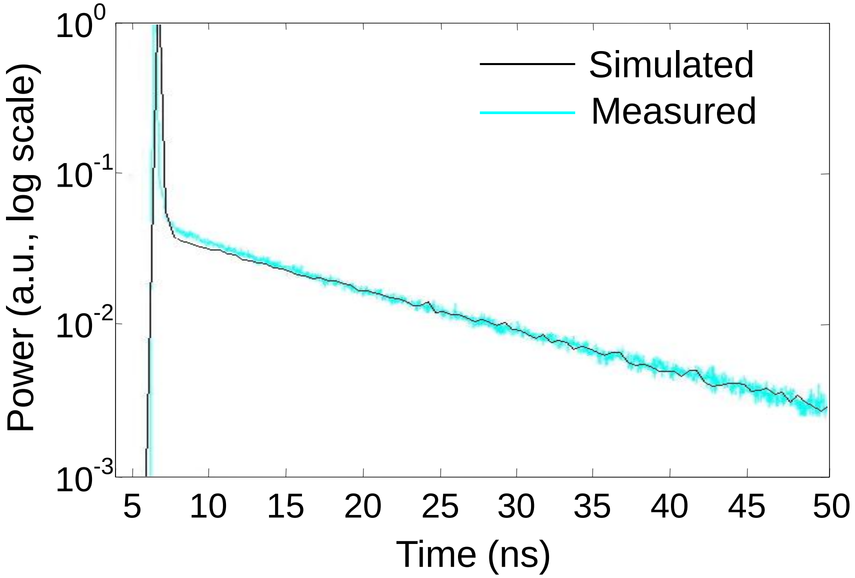
Conclusions and future work

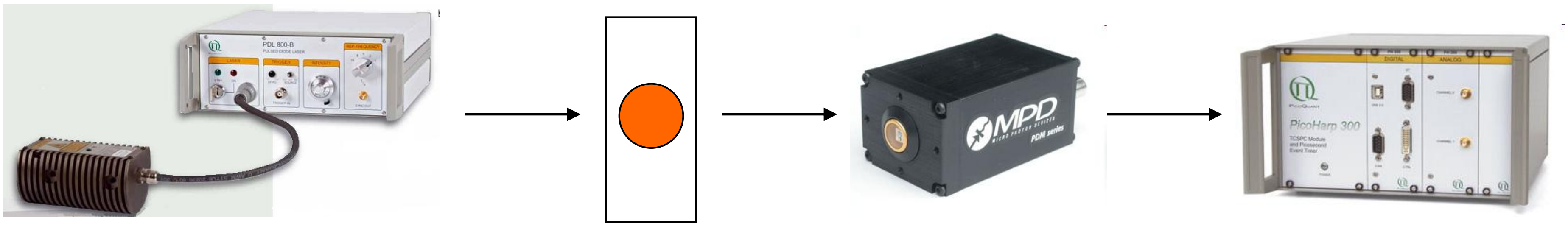
Test 1
Time-correlated single photon counting

Experimental setup

- PicoQuant pulsed laser
- Quantum dots in a micro cavity
- MPD® SPAD
- PicoHarp 300 TCSPC module

Results



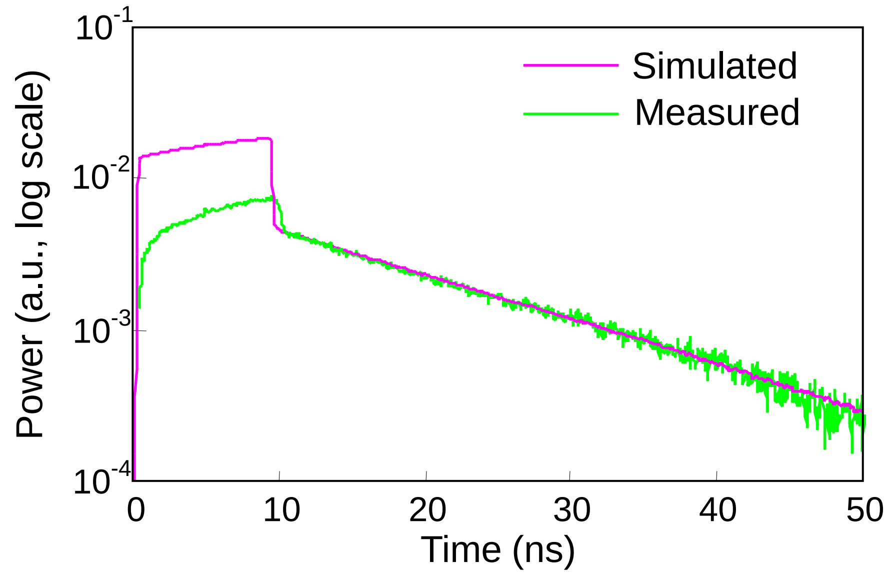


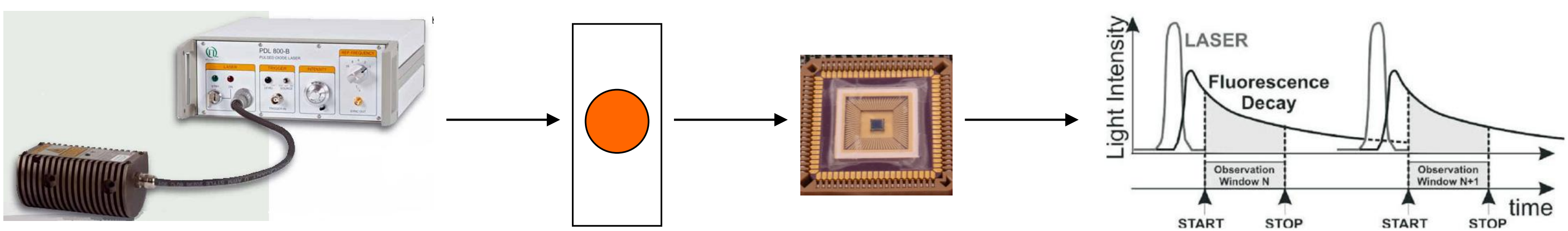
Modified time-gating

Modified time-gating

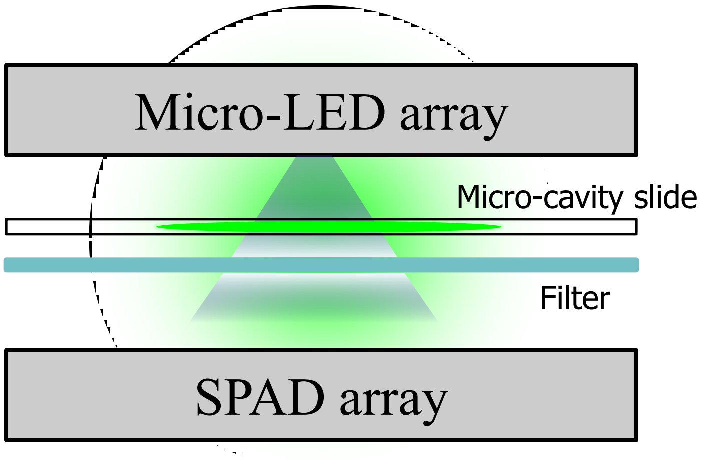
- PicoQuant pulsed laser
- Quantum dots in a micro cavity
- FBK SPAD
- Modified time-gating

Results





Two-chip micro-system



- Blue micro-LED
- Quantum dots
- Semrock filter
- CMOS SPAD
- Becker & Hickl TCSPC module

- Simulation
  - Single pixel in SPAD array and nearest LED from LED array
  - Passive quenching
  - Backward simulation type
- Experiment 1: different LED pulse width
- Experiment 2: different setup

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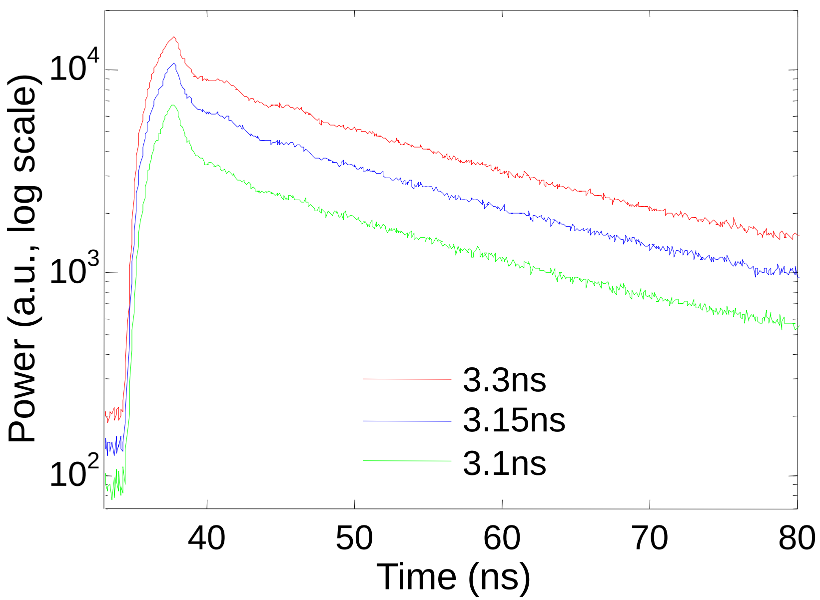
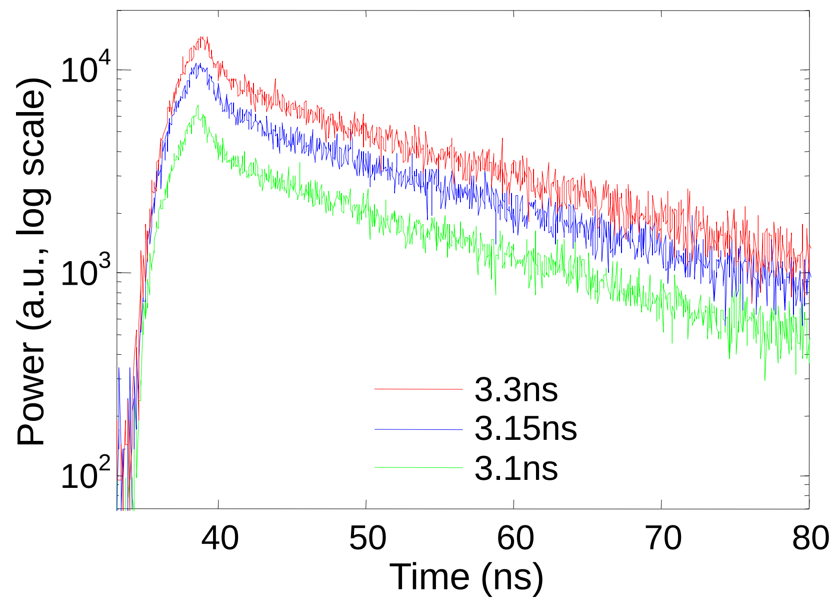
Test 2
Different pulse width

Experiment



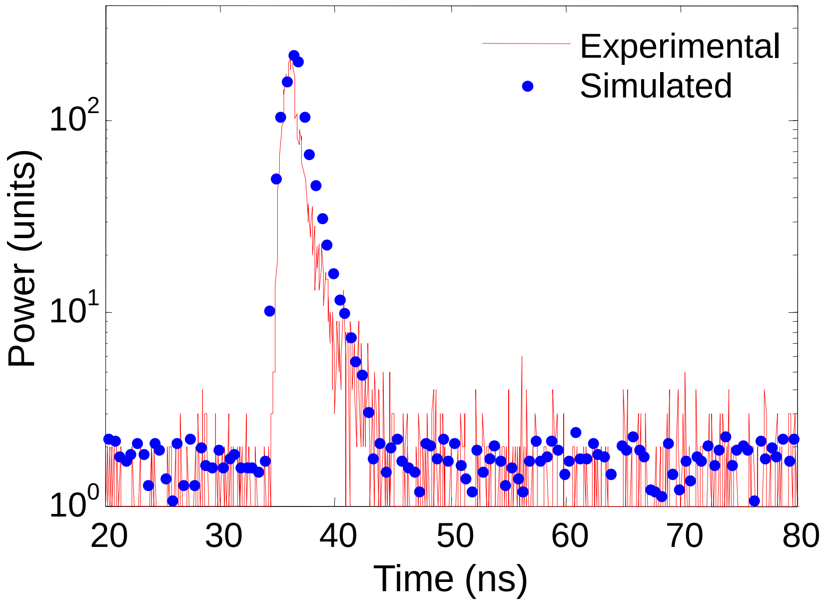
Simulation



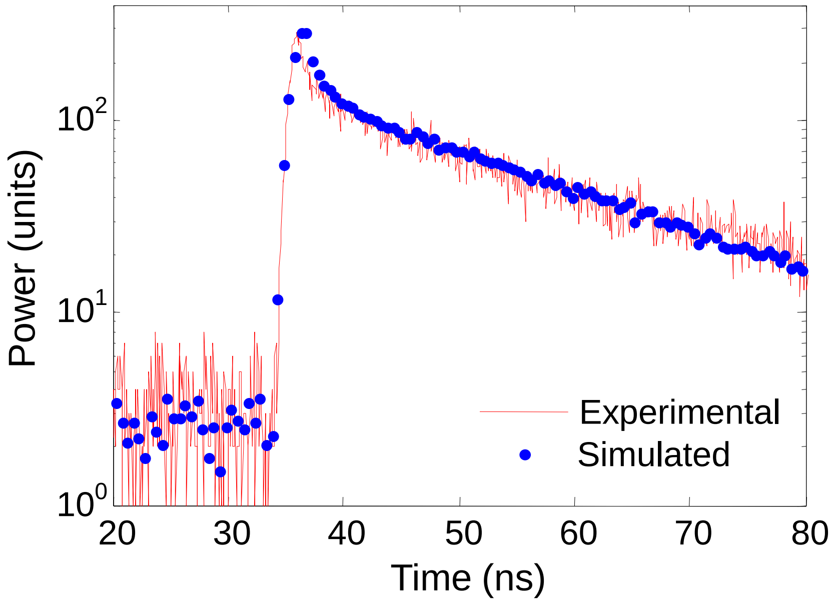
- **Conclusions:**
  - The simulated and measured results are in good agreements

Different setup

LED + Filter + SPAD

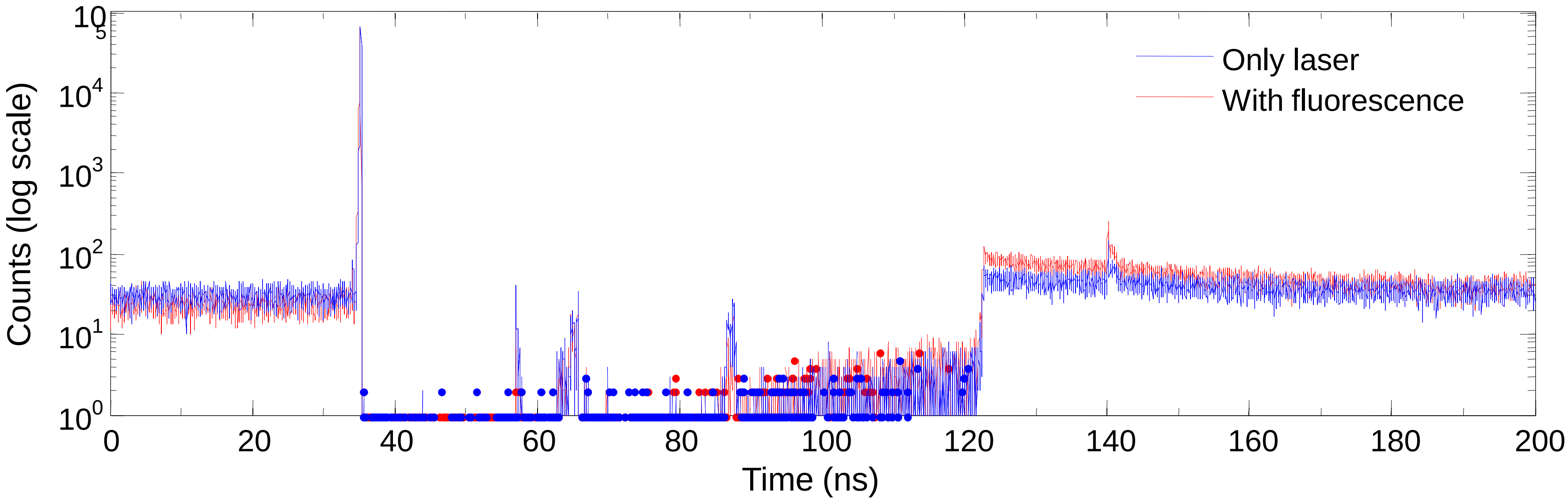


LED + Filter + Fluo + SPAD



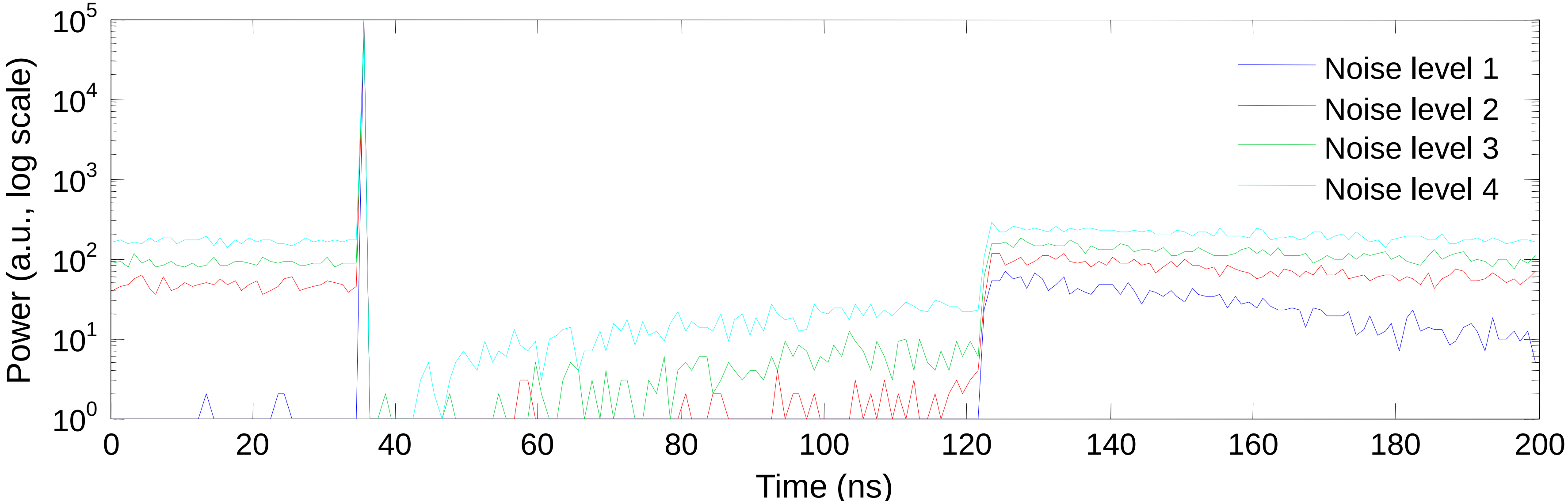
- **Conclusions:**
  - The simulated and measured results are in good agreements except the peak area
  - The mismatch can be explained by the lack of exact LED time curve used in simulation

Pile-up effect investigation



- **Picoquant pulsed laser 1MHz**
- **Quantum dots in a micro cavity**
- **MPD® single SPAD**
- **PicoHarp 300 TCSPC module**

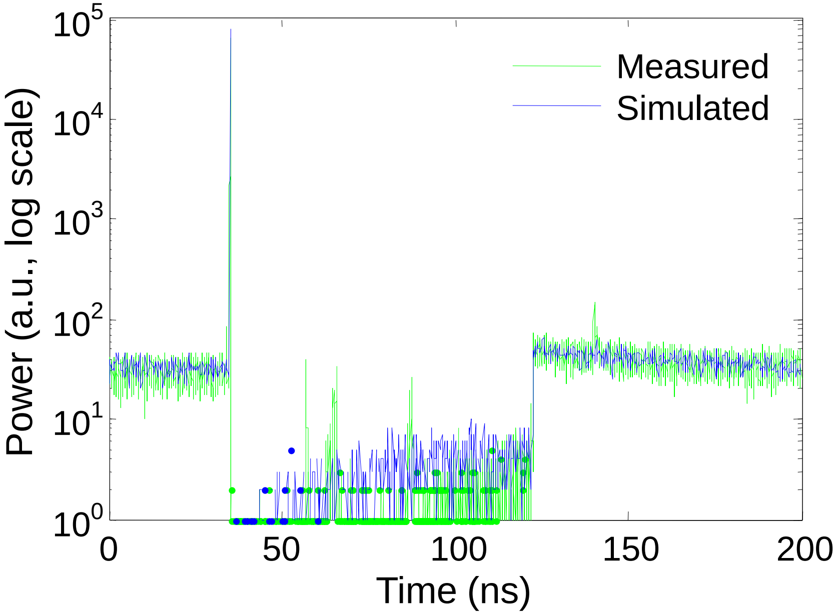
Pile-up with different ambient light



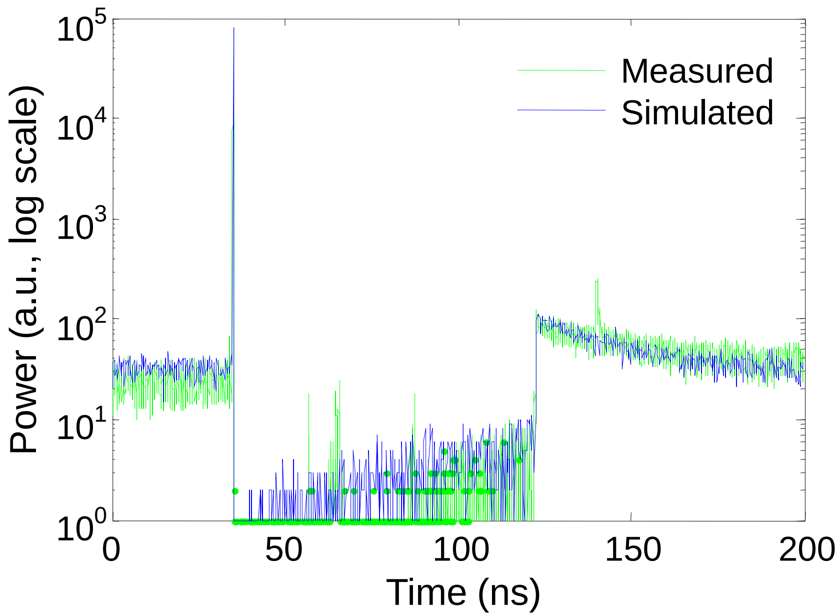
- **Simulation of the same setup**
- **Noise level 1 < Noise level 2 < Noise level 3 < Noise level 4**
- **Conclusions:**
  - the increasing number of counts near the end of TCSPC dead time is due to noise detection before laser pulse
  - higher noise level after TCSPC dead time is afterpulsing

Complete simulation of pile-up

Laser light



Fluorescence decay

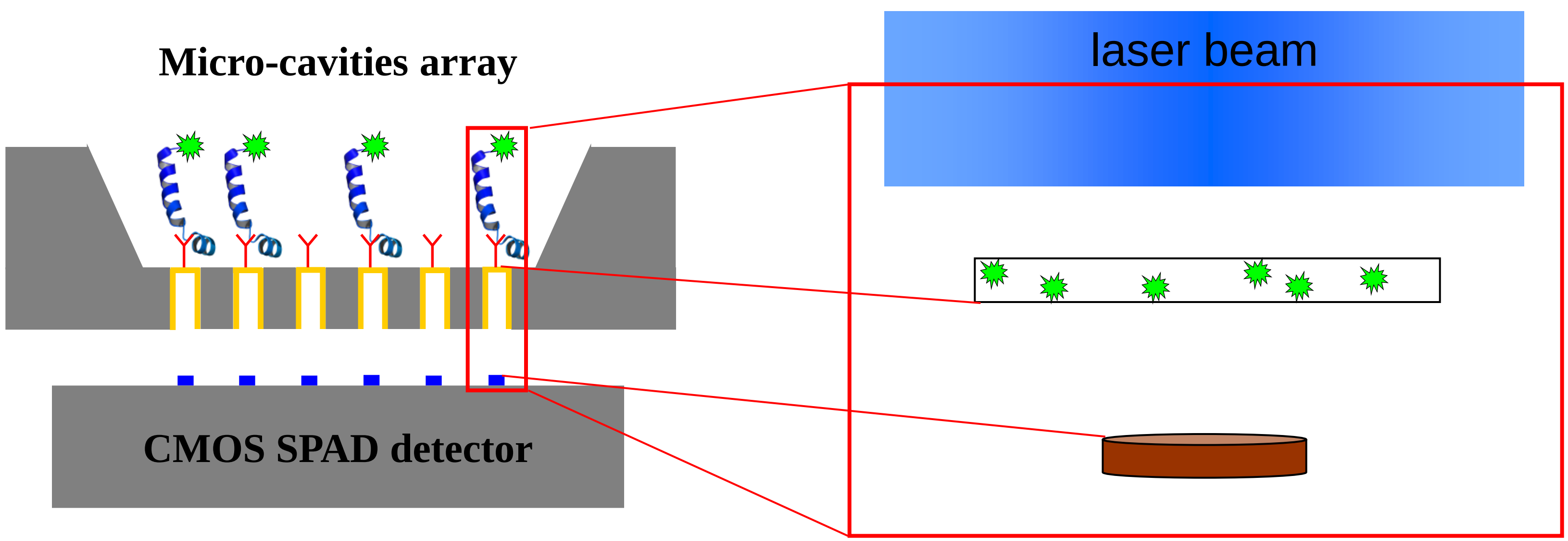


- **Conclusions:**
  - These experiments demonstrated the ability of the proposed simulation model to help in explanation of some unexpected results of the real measurements.

Microarray system

Real system

Simulated analog

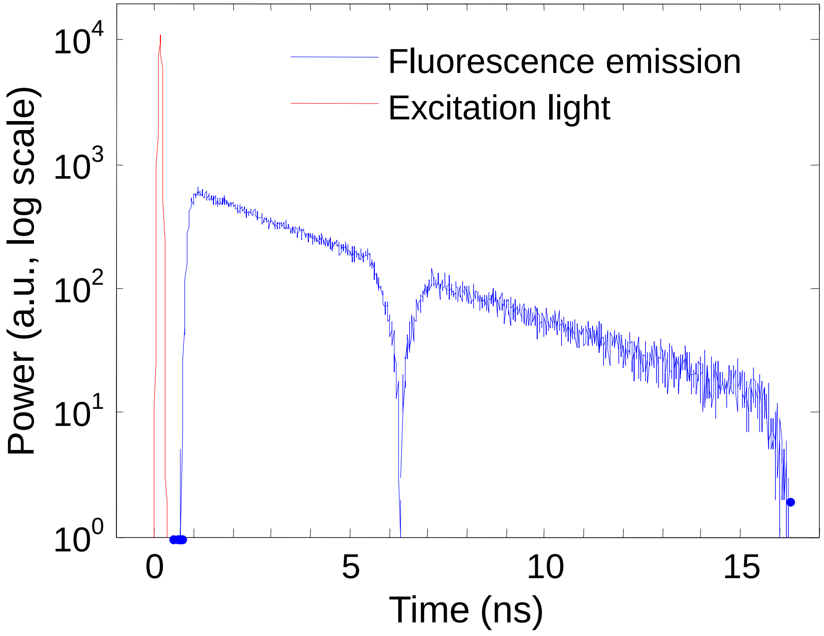


- How perfect is the time-filter in comparison to the conventional optical filter?
- What is the influence of the "noisy" pixels on the lifetime estimation?
- How does the OW width affect the lifetime estimation?

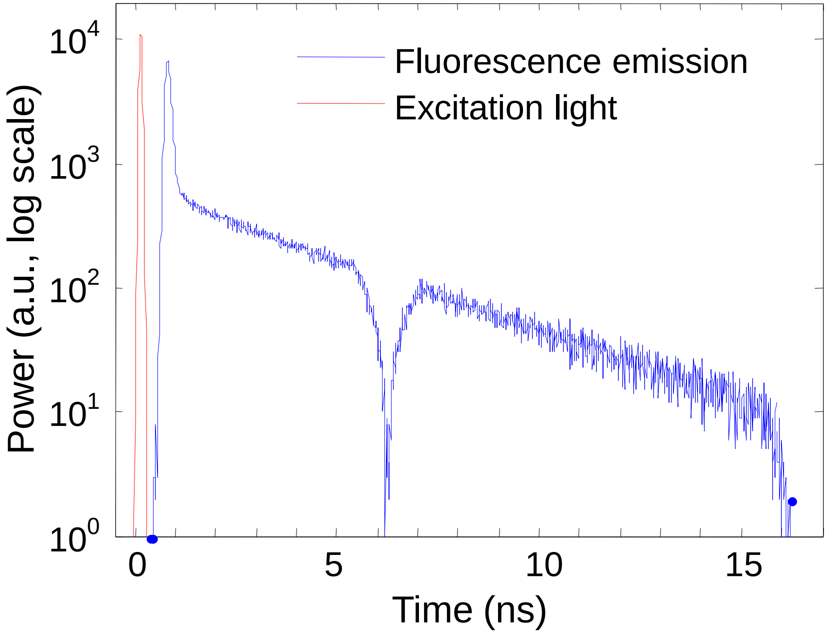
Test 4

Time filtering

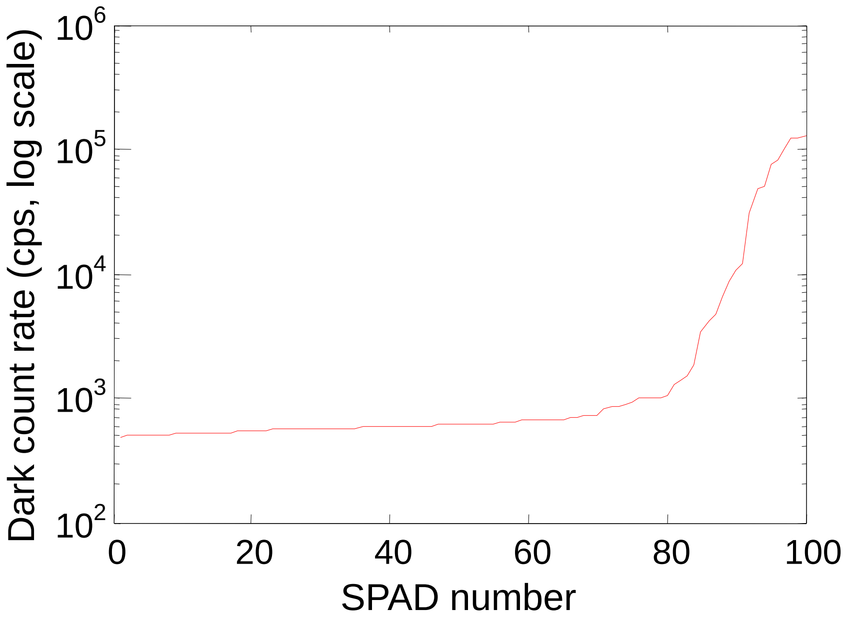
0.3 ns SPAD switched-off time



0.25 ns SPAD switched-off time



Noise characteristic



- **80% pixels ~ 900Hz**
- **20% pixels up to 150kHz**
- **Two time intervals**
  - 10 ns and 10 ns
  - 6 ns and 6 ns
- **Calculated:**
  - Lifetime per pixel
  - Cumulative lifetime

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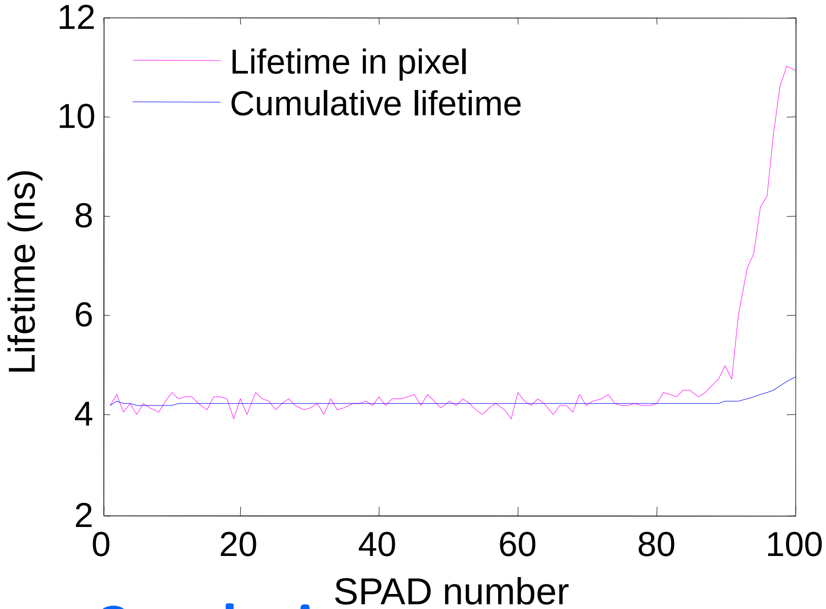
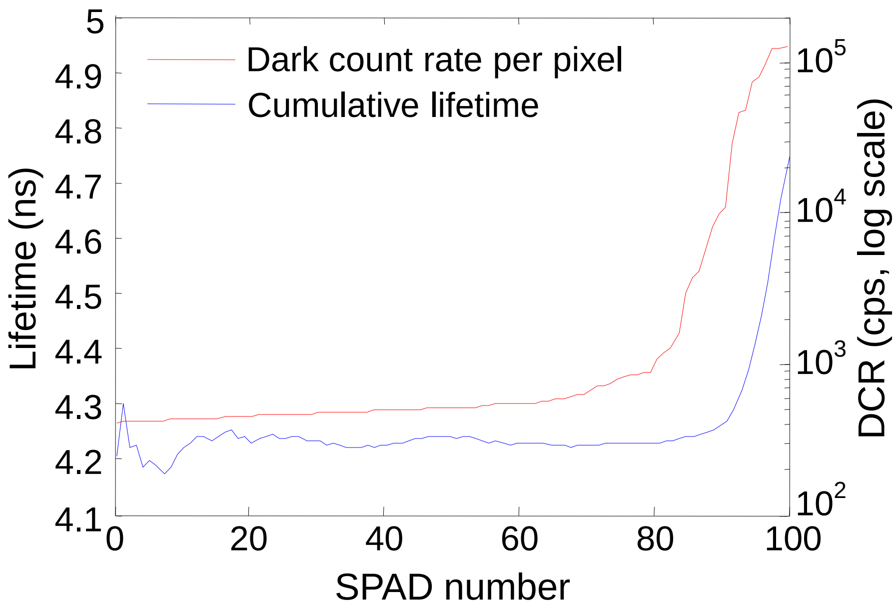
Test 4

10 ns time intervals

Per pixel and cumulative lifetime



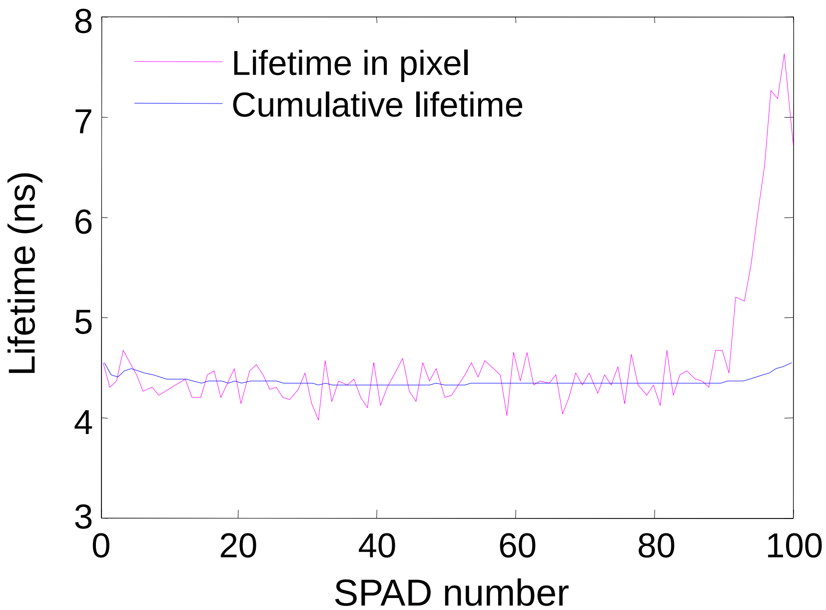
Cumulative lifetime and noise characteristic



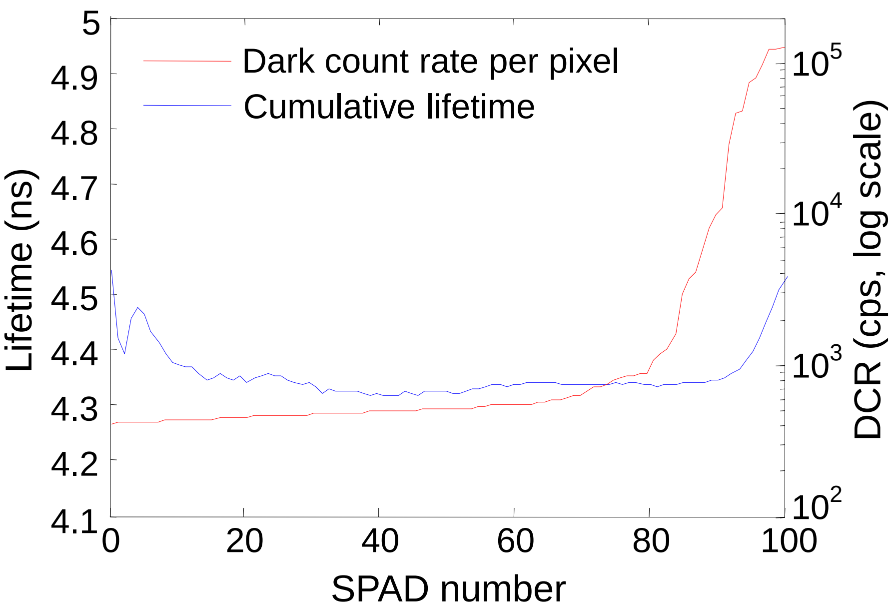
- **Conclusions:**
  - Cumulative life time has smaller statistical noise
  - In the case of very high noise per pixel cumulative lifetime starts increasing
  - It is better not to include information from noisy pixels to lifetime extraction

6 ns time intervals

Per pixel and cumulative lifetime



Cumulative lifetime and noise characteristic



- **Conclusions:**
  - In case of non optimal time interval width the extracted lifetime has worth accuracy
  - Using the average of several pixel improves the accuracy of lifetime extraction

Road Map

Introduction

Proposed model

Validation of the model

Conclusions and future work

Summary

- A simulator of typical fluorescence measurement experiment has been designed
- It has been verified by real experiments
- The qualitative results are in a very good agreement to the real experiments
- The quantitative simulation is within the limits of accuracy of input parameters and made assumptions

Conclusions

The created model can…

- **explain unexpected experimental results**
- **predict the results of real and planned experiments**
- **enable the preproduction-stage investigation of SPAD detectors**

Future work

- **Advanced models of experimental components**
  - Fluorescence is not a point source
  - Lenses simulation
- **More complex biological system**
  - Energy transfer (for FRET)
- **Optimization algorithm**
- **Conversion to a compiled programming language**

Thank you