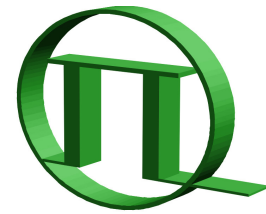
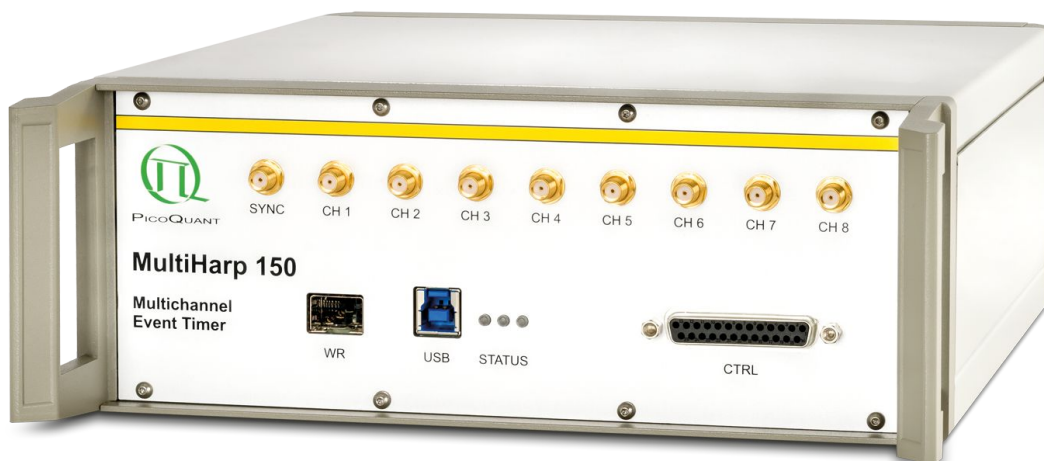


MultiHarp 150



PICOQUANT GmbH
Unternehmen für optoelektronische
Forschung und Entwicklung

Multichannel Time–Correlated Single Photon Counting System and Time Tagger with USB Interface



User's Manual and Technical Data

Version 1.1.0.0

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Acknowledgements

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When the MultiHarp software is used under Linux it uses Libusb to access the MultiHarp USB devices. Libusb is licensed under the LGPL which allows a fairly free use even in commercial projects. For details and precise terms please see <http://libusb.info>. In order to meet the license requirements a copy of the LGPL as applicable to Libusb is provided as part of the MultiHarp software distribution media. The LGPL does not apply to the MultiHarp software as a whole.

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1. Introduction

While intensity based fluorescence spectroscopic investigations have been fairly common for a long time, extracting additional temporal information from quantum systems via pulsed excitation and time-resolved detection is a relatively new and powerful technique. The temporal analysis can reveal information about the emitter that is not available from spectral data alone. This is why time-resolved analysis of (typically laser induced) fluorescence by means of Time–Correlated Single Photon Counting (TCSPC) has gained in importance over the recent years. For instance, in life sciences the difference in the fluorescence decay times of fluorophores provides a powerful discrimination feature to distinguish molecules of interest from background or other species. This has made the technique very interesting for sensitive analysis, even down to the single molecule level. The same mechanisms are applicable in quantum optics, e.g., when defect centers in diamond are observed.

The acquisition of fluorescence decay curves by means of TCSPC provides resolution and sensitivity that cannot be achieved with other methods. In practice it is done by histogramming arrival times of individual photons over many excitation and fluorescence cycles. The arrival times recorded in the histogram are relative times between laser excitation and corresponding fluorescence photon arrival (start / stop times) ideally resolved down to a few picoseconds. The resulting histogram represents the fluorescence decay. Although fluorescence lifetime analysis is a great field of application for the MultiHarp 150, it is in no way restricted to this task. Other important applications are e.g. quantum optics, Quantum Cryptography (QC) Time–Of–Flight (TOF) and Optical Time Domain Reflectometry (OTDR) as well as any kind of coincidence correlation.

The MultiHarp 150 is a cutting edge TCSPC system with USB interface. Its new integrated design provides a flexible number of input channels at reasonable cost and allows innovative measurement approaches. The timing circuits allow high measurement rates up to 78 million counts per second (Mcps) with an excellent time resolution of 80 ps and a deadtime of 650 ps. The modern USB 3.0 interface provides very high throughput as well as 'plug and play' installation. The input triggers are adjustable for a wide range of input signals providing programmable level triggers for both negative and positive going signals. These specifications qualify the MultiHarp 150 for use with most common single photon detectors such as Single Photon Avalanche Diodes (SPADs) and Photomultiplier Tube (PMT) modules (via preamplifier). Depending on detector and excitation source the width of the overall Instrument Response Function (IRF) can be as small as 200 ps FWHM. The MultiHarp 150 can be purchased with 4, 8 or 16 timing inputs and one synchronization (sync) input. The use of these inputs is very flexible. In fluorescence lifetime applications the sync channel is typically used as a synchronization input from a laser. The other inputs are then used for photon detectors. In coincidence correlation applications all inputs including the sync input can be used for photon detectors.

The MultiHarp 150 can operate in various modes to adapt to different measurement needs. The standard histogram mode performs real-time histogramming in on-board memory. Two different Time–Tagged–Time–Resolved (TTTR) modes allow recording of each photon event on separate, independent channels, thereby providing unlimited flexibility in off-line data analysis such as burst detection and time-gated or lifetime weighted Fluorescence Correlation Spectroscopy (FCS) as well as picosecond coincidence correlation, using the individual photon arrival times. The MultiHarp 150 is furthermore supported by a variety of accessories such as pre-amplifiers, signal adaptors and detector assemblies from PicoQuant. A significant novel feature of the MultiHarp 150 is support for White Rabbit, allowing time transfer and synchronization with sub-ns accuracy (see https://en.wikipedia.org/wiki/The_White_Rabbit_Project).

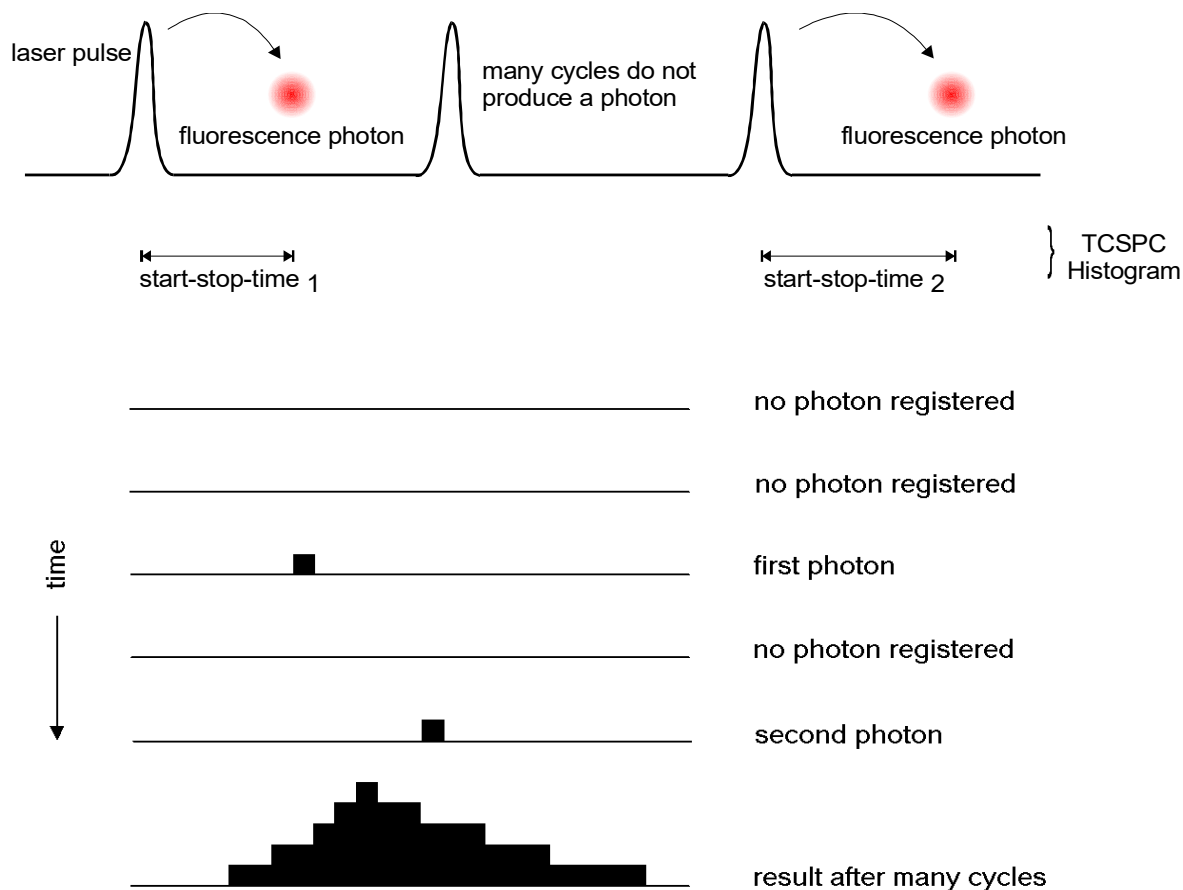
The MultiHarp software runs on current Windows PC platforms including the x64 editions. It provides functions for setting measurement parameters, displaying measurement results, loading and saving of measurement parameters and decay curves. Important measurement characteristics such as count rate, count maximum and position, histogram width (FWHM) are displayed continuously. Data can conveniently be exported via the clipboard, e.g. for immediate processing by the EasyTau 2 Fluorescence Decay Fit Software. Furthermore, a programming library (DLL) enables users to write custom data acquisition programs for the MultiHarp in virtually all popular programming environments. There is also a library version for Linux (Intel processor architecture only) which is fully compatible with that for Windows so that applications can easily be ported across the two platforms.

For details on the Time–Correlated Single Photon Counting method, please read the next section as well as our TechNote on TCSPC and consult the literature referenced at the end of section 2.4. Experienced users of the method should be able to work with the MultiHarp 150 straight away. Nevertheless, we recommend carefully reading sections 3.3 and 3.4 on hardware and software installation to avoid damage. Later, the comprehensive online-help function of the MultiHarp software will probably let the manual gather dust on the shelf.

2. Primer on Time–Correlated Single Photon Counting

In order to make use of a powerful analysis tool such as time–resolved fluorescence spectroscopy, one must record the time dependent intensity profile of the emitted light. While in principle, one could attempt to record the time decay profile of the signal from a single excitation / emission cycle, there are practical problems that prevent such a simple solution in most cases. First of all, the decay to be recorded is very fast. Typical fluorescence from organic fluorophores lasts only a few hundred picoseconds to some hundred nanoseconds. In order to recover fluorescence lifetimes as short as e.g., 500 ps, one must be able to resolve the recorded signal at least to such an extent, that the exponential decay is represented by enough sample points in time. This means that the required transient recorder would have to sample at very high rates. This is hard to achieve with ordinary electronic transient recorders of reasonable dynamic range. Secondly, the light available may simply be too weak to sample an analog time decay. Indeed the signal may consist of just single photons per excitation / emission. This is typically the case for single molecule experiments or work with minute sample volumes / concentrations. Then the discrete nature of the signal itself prohibits analog sampling. Even if one has more than just a single molecule and some reserve to increase the excitation power to obtain more fluorescence light, there will be limits, e.g. due to collection optic losses, spectral limits of detector sensitivity or photo–bleaching at higher excitation power. The solution is Time–Correlated Single Photon Counting (TCSPC). By using periodic excitation (e.g., from a laser) it is possible to extend the data collection over multiple excitation/emission cycles and one can then reconstruct the single cycle decay profile from single photon events collected over many cycles.

The TCSPC method is based on the repetitive, precisely timed registration of single photons of e.g., a fluorescence signal. The reference for the timing is the corresponding excitation pulse. A single photon detector such as a Photo Multiplier Tube (PMT) or a Single Photon Avalanche Photodiode (SPAD) is used to capture the fluorescence photons. Provided that the probability of registering more than one photon per cycle is low, the histogram of photon arrivals per time bin represents the time decay one would have obtained from a single shot time–resolved analog recording. The precondition of single photon probability can (and must) be met by attenuating the light level reaching the sample if necessary. If the single photon probability condition is met, there will actually be no photons registered in many of the excitation cycles. The diagrams below illustrate how the histogram is formed over multiple cycles.



The histogram is collected in a block of memory, where one memory cell holds the photon counts for one corresponding time bin. These time bins are often (historically) referred to as “time channels”. In practice, the registration of one photon involves the following steps: first, the time difference between the photon event and the corresponding excitation pulse must be measured. For this purpose both optical signals are converted to electrical signals. For the fluorescence photon this is done via the single photon detector mentioned previously. For the excitation pulse it may be done via another detector if there is no electrical sync signal supplied by the laser directly. Obviously, all conversion to electrical pulses must preserve the precise timing of the signals as accurately as possible. The actual time difference measurement is done by means of fast electronics which provide a digital timing result. This digital timing result is then used to address the histogram memory so that each possible timing value corresponds to one memory cell or histogram channel. Finally the addressed histogram cell is incremented. All steps are carried out by fast electronics so that the processing time required for each photon event is as short as possible. When sufficient counts have been collected, the histogram memory can be read out. The histogram data can then be used for display and e.g., fluorescence lifetime calculation. In the following we will expand on the various steps involved in the method and related issues of importance.

2.1. Count Rates and Single Photon Statistics

It was mentioned that in the past it was necessary to maintain a low probability of registering more than one photon per cycle. This condition ensured that the histogram of photon arrivals represents the time decay one would have obtained from a single shot time-resolved analog recording. The reason for this is briefly the following: Due to dead times of detector and electronics lasting at least some tens of nanoseconds after a photon event, historical TCSPC systems could register only one photon per excitation / emission cycle. If then the number of photons occurring in one excitation cycle were typically > 1 , the system would very often register the first photon but miss the following one or more. This would lead to an over-representation of early photons in the histogram, an effect called ‘pile-up’. This leads to distortions of the fluorescence decay, typically making the fluorescence lifetime appear shorter. It was therefore crucial to keep the probability of cycles with more than one detected photon sufficiently low, unless one tolerates some error and/or corrects for it in data analysis.

To quantify the “safe” count rate limit for traditional TCSPC, one had to set acceptable error limits and apply some mathematical statistics. For practical purposes one would apply a rule of thumb: In order to maintain single photon statistics, on average only one in 20 to 100 excitation pulses should generate a count at the detector. In other words: the average count rate at the detector should be at most 1 % to 5 % of the excitation rate. Using e.g., a diode laser from PicoQuant's PDL Series, pulsed at 80 MHz repetition rate, the average detector count rate would then not exceed 4 Mcps. Typically one wants high count rates, in order to acquire fluorescence decay histograms quickly. This is of particular importance where dynamic lifetime changes or fast molecule transitions are to be studied or where large numbers of lifetime samples must be collected (e.g., in image scanning). This is why high laser rates (such as 40 or 80 MHz for the PDL Series) are important. PMTs can safely handle TCSPC count rates of over 10 Mcps. Most actively quenched SPADs may also operate up to 10 Mcps but some types suffer resolution degradation when operated too fast. Old NIM based TCSPC electronics could only handle a maximum of 50 to 500 kcps, while newer integrated TCSPC electronics typically reach count rates of 5 to 40 Mcps. With the MultiHarp 150 and modern Hybrid Photodetectors (HPD) count rates up to 78 Mcps can be collected.

It is worth noting that the photon arrival times are typically random so that there can be bursts of high count rate and periods of low count rates. Bursts may exceed the average rate. The average (sustained) rate the instrument can deal with is important when losses cannot be tolerated, notably in time tagging applications. Due to the high bandwidth of USB 3.0 the MultiHarp 150 can handle sustained rates as high as 80 Mcps. When comparing count rates considered here and elsewhere please pay attention to the details. The specifications for TCSPC systems often interpret their maximum count rates differently. This is why in this context dead-time is also of interest. It describes the time the system cannot register photons while it is processing a previous photon event. The term is applicable both to detectors and electronics. The MultiHarp 150 has an extremely short dead time of about 650 ps, imposing the smallest losses among comparable instruments today. The short dead time of the MultiHarp 150 together with its multi-stop capability now allows measurement scenarios where the classic pile-up limit is no longer critical. This is of special interest for very fast FLIM measurements and PicoQuant's concept of rapidFLIM exploits the idea. Using an HPD (e.g., PicoQuant PMA Hybrid) that permits count rates as high as 80 Mcps one can obtain FLIM images at unprecedented speed. While classic pile-up due to dead time is no longer an issue, one must now consider the effect of pulse-pile-up. This effect occurs when photons arrive at the detector with very short temporal spacing, such that the detector can no longer produce separate output pulses. This merging of detector pulses leads to another type of artifact in the decay histograms. A numerical correction of these artifacts during data analysis ensures that lifetimes and amplitudes are still obtained correctly for very fast quantitative measurements. See section 2.5 or www.picoquant.com for related publications.

2.2. Timing Resolution

The most critical component for the timing resolution in TCSPC measurements is usually the detector. However, in contrast to analog transient recording, the time resolution of TCSPC is not limited by the impulse response of the detector. Only the timing accuracy of registering a photon determines the resolution. This is limited by the timing uncertainty that the detector introduces in the conversion from a photon to an electrical pulse. This timing error or uncertainty can be as much as ten times smaller than the detector's pulse response. The timing uncertainties are usually quantified by specifying the rms error (standard deviation) or the Full Width at Half Maximum (FWHM) of the timing distribution or instrument response function (IRF). Note that these two notations are related but not identical¹. Micro channel plate PMTs, can achieve timing uncertainties as small as 25 ps FWHM. Lower cost PMTs or SPADs may introduce uncertainties of 50 to 500 ps FWHM, HPDs lie in between with typical uncertainties of 50..150 ps FWHM.

The second most critical source of IRF broadening in fluorescence lifetime measurements with TCSPC is usually the excitation source. While many laser sources can provide sufficiently short pulses, it is also necessary to obtain an electrical timing reference signal (sync) for comparison with the fluorescence photon signal. The type of sync signal that is available depends on the excitation source. With gain switched diode lasers (e.g., PDL 800-D) a low jitter electrical sync signal is readily available. The sync signal used here is typically a narrow negative pulse of -800 mV into 50 Ω (NIM standard). The sharp falling edge is synchronous with the laser pulse (< 3 ps rms jitter for the PDL 800-D). With other lasers (e.g., Ti:Sa) a second detector must be used to derive a sync signal from the optical pulse train. This is commonly done with a fast photo diode (APD or PIN diode). The light for this reference detector must be derived from the excitation laser beam e.g., by means of a semi-transparent mirror. The reference detector must be chosen and set up carefully as it contributes to the overall timing error.

Another source of timing error is the timing jitter of the electronic components used for TCSPC. This is caused by the finite rise / fall-time of the electrical signals used for the time measurement. At the trigger point of comparators, logic gates etc., the amplitude noise (thermal noise, interference etc.) always present in these signals is transformed to a corresponding timing error (phase noise). However, the contribution of the electronics to the total timing error is usually small. For the MultiHarp 150 random jitter is less than 85 ps rms.

Generally, it is always a good idea to keep electrical noise pick-up low in all system components. Uncorrelated electrical noise will cause just random jitter and IRF broadening but correlated noise can cause even more disturbing artifacts. This is why signal leads should be properly shielded coax cables, and strong sources of electromagnetic interference should be kept away from the TCSPC detector and electronics.

The contribution of the time spread introduced by the individual components of a TCSPC system to the total IRF strongly depends on their relative magnitude. Strictly speaking, the overall IRF is the convolution of all component IRFs. An estimate of the overall IRF width, assuming independent noise sources, can be obtained from the geometric sum of the individual components as an rms figure according to statistical error propagation laws:

$$e_{system} = \sqrt{\sum e_{component}^2}$$

As the individual contributions are squared, the total will be dominated by the largest component. It is therefore of little value to improve a component that is already relatively good. If e.g., the detector has an IRF width of 200 ps FWHM, shortening the laser pulse from 50 ps to 40 ps will have practically no effect. Although it is difficult to specify a general lower limit on the fluorescence lifetime that can be measured by a given TCSPC instrument, as a rule of thumb, one can assume that under favourable conditions lifetimes down to 1 / 10 of the IRF width (FWHM) can still be recovered via deconvolution.

A final time-resolution related issue worth noting here is the bin width of the TCSPC histogram. As outlined above, the analog electronic processing of the timing signals (detector, amplifiers, etc.) creates a continuous distribution around any true time value. In order to form a histogram, at some point the timing results must be quantized. This quantization introduces further error, if chosen too coarse. The quantization step width (i.e. the resolution) should therefore be small compared to the IRF width. As a minimum sampling frequency, from the point of view of information theory, one would assume the Nyquist frequency. That is, the signal should be sampled at least at twice the highest frequency contained in it. For practical purposes one may wish to exceed this limit where possible, but there is usually little benefit in sampling the histogram at resolutions higher than 1 / 10 of the overall IRF width of the analog part of the system.

¹ In case of a Gaussian distribution the r.m.s deviation corresponds to the standard deviation σ and FWHM $\approx 2,35 \sigma$.

2.3. Photon Counting Detectors

2.3.1. Photomultiplier Tube (PMT)

A PMT consists of a light-sensitive photo cathode that generates electrons when exposed to light. These electrons are directed onto a charged electrode called dynode. The collision of the electrons with the dynode produces additional electrons. Since each electron that strikes the dynode causes several electrons to be emitted, there is a multiplication effect. After further amplification by multiple dynodes, the electrons are collected at the anode of the PMT and output as a current. The current is directly proportional to the light intensity striking the photo cathode. Because of the multiplicative effect of the dynode chain, the PMT is a photo electron amplifier with high sensitivity and remarkably low noise. The high voltage driving the tube may be varied to change the sensitivity of the PMT. Current PMTs have a wide dynamic range, i.e. they can also measure relatively high levels of light. Furthermore, they are very fast, so that rapid successive events can be reliably monitored. One photon on the photo cathode can produce a short output pulse containing millions of photoelectrons. PMTs can therefore be used as single photon detectors. In photon counting mode, individual photons that strike the photo cathode of the PMT are registered. Each photon event gives rise to an electrical pulse at the output. The number of pulses, or counts per second, is proportional to the light impinging upon the PMT. As the number of photon events increase at higher light levels, it will become difficult to differentiate between individual pulses and the photon counting detector's behavior will become non-linear. This usually occurs between 1 to 10 Mcps, depending on the detector design. Similarly, in TCSPC applications, individual photon pulses may merge into one as the count rate increases. This leads to pulse pile-up and distortions of the collected histograms.

The timing uncertainty between photon arrival and electrical output (transit time spread) is usually small enough to permit time-resolved photon counting at a sub-nanosecond scale. In single photon counting mode the tube is typically operated at a constant high voltage where the PMT is most sensitive.

PMTs usually operate within the blue to red regions of the visible spectrum, with greatest quantum efficiency in the blue-green region, depending upon photo-cathode materials. Typical quantum efficiencies are about 25 %. For spectroscopy experiments in the ultraviolet / visible / near infrared region of the spectrum, a PMT is very well suited.

Because of noise from various sources in the tube, the output of the PMT may contain pulses that are not related to the light input. These are referred to as dark counts. The detection system can to some extent reject these spurious pulses by means of electronic discriminator circuitry. This discrimination is based on the probability that some of the noise generated pulses (those from the dynodes) exhibit lower signal levels than pulses from a true photon event. Thermal emission from the cathode that undergoes the full amplification process can usually not be suppressed this way. In this case cooling of the detector is more helpful.

2.3.2. Micro Channel Plate PMT (MCP)

A Micro Channel Plate PMT consists of an array of glass capillaries (5–25 μm inner diameter) whose insides are coated with an electron-emissive material. The capillaries are biased at a high voltage. Like in a PMT, an electron that strikes the inside wall of one of the capillaries creates an avalanche of secondary electrons. This cascading effect creates a gain of 10^3 to 10^6 and produces a current pulse at the output. Due to the narrow and well defined electron path inside the capillaries, the transit time spread of the output pulses is much reduced compared to a normal PMT. The timing jitter of MCPs is therefore sufficiently small to perform time-resolved photon counting on a picosecond scale, usually outperforming PMTs. Good but also expensive MCPs can achieve timing uncertainties as low as 25 ps. In this respect, microchannel plates are a good match for the MultiHarp 150 but they are quite limited in permitted count rate and provide lower sensitivity towards the red end of the spectrum compared to suitably optimized SPADs.

2.3.3. Single Photon Avalanche Photo Diode (SPAD)

Avalanche Photo Diodes (APDs) are semiconductor devices, usually restricted to operation in the visible to infrared parts of the spectrum. Generally, APDs may be used for ultra-low light detection (optical powers $< 1 \text{ pW}$), and can also be used as photon-counters in the so-called "Geiger" mode (biased slightly above the breakdown voltage). In the latter case, a single photon may trigger an avalanche of about 10^8 carriers. In this mode the device can be used as a detector for photon counting with very accurate timing of the photon arrival. In this context they are also referred to as Single Photon Avalanche Photo Diodes (SPAD). Selected devices with small active surfaces may achieve timing accuracies down to 50 ps, but are usually hard to align and difficult to focus into. SPADs are sometimes noisier than PMTs, but can have a greater quantum efficiency especially towards

the red side of the spectrum. Maximum quantum efficiencies are about 70 %. Such sensitive devices provide a timing accuracy of ~400 ps. Commercial modules are thermoelectrically cooled for low dark count rate and deliver pre-shaped TTL pulses. They are the most common detectors for applications where NIR sensitivity is important, e.g., single molecule detection. To achieve the specified timing accuracy, exact focusing into the center of the active area is necessary. Other SPAD designs such as the PDM family from Micro Photon Devices have the benefit of much better timing resolution and robustness, however, at the expense of a lower sensitivity at the red end of the spectrum.

2.3.4. Other and Novel Photon Detectors

The field of photon detectors is still evolving. Recent developments include so called silicon PMTs, Hybrid Photo Detectors (HPDs), superconducting nanowire detectors and APDs with sufficient gain for single photon detection in analog mode. Each of these detectors have their specific benefits and shortcomings. Only a very brief overview will be given here.

Silicon PMTs are essentially arrays of SPADs, all coupled to a common output. This has the benefit of creating a large area detector that can even resolve photon numbers. The drawback is increased dark count rate and reduced timing accuracy.

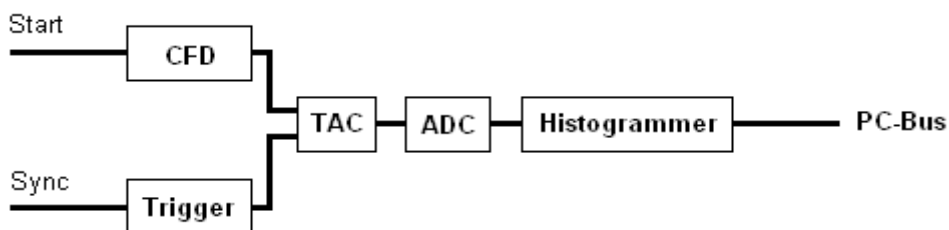
HPDs make use of a combination of a PMT-like front end followed by an APD structure. The benefits are good timing performance and virtually zero afterpulsing while the need for very high voltage is a disadvantage. PicoQuant's PMA Hybrid series include the high voltage in an easy-to-use package.

Superconducting nanowires single photon detectors (SNSPDs or SSPDs) routinely achieve excellent timing performance (<30 ps jitter) and high sensitivity from the visible to the near infrared with overall system detection efficiencies in excess of 90%. They operate at cryogenic temperatures, typically between 0.8 K and 4 K, which incurs cost, large footprint and power consumption and makes them less practical for applications where these parameters are of concern.

Another class of potentially interesting detectors which have recently emerged, are APDs with very high gain. In combination with an electronic amplifier they have been shown to detect single photons. As opposed to Geiger mode, this avoids afterpulsing and allows for very fast counting rates. The disadvantage is a high dark count rate, currently too high for any practical TCSPC application.

2.4. Principles Behind the TCSPC Electronics

For introductory purposes it is worth to look first at the design of historical TCSPC systems. They consist of the following building blocks:



The Constant Fraction Discriminator (CFD) is used to extract precise timing information from the electrical detector pulses that may vary in amplitude. This way the overall system IRF may be tuned to become narrower and some of the random background signal can be suppressed. The same could not be achieved with a simple level trigger (comparator). Especially with PMTs, constant fraction discrimination is important, because their pulse amplitudes vary significantly. In particular pulses originating from random electrons generated at the dynodes of the PMT can be suppressed because their avalanches had less time to amplify and their corresponding output pulses are small.

The principle of a classic CFD is the comparison of the original detector signal with an amplified, inverted and delayed version of itself. The signal derived from this comparison changes its polarity exactly when a constant fraction of the detector pulse height is reached. The zero crossing point of this signal is therefore suitable to derive a timing signal independent from the amplitude of the input pulse. In practice the comparison is done by a summation. The timing is done by a subsequent threshold trigger of the sum signal using a settable level, the so called zero cross trigger. Newer CFD designs achieve the same objective by differentiating the input signal and

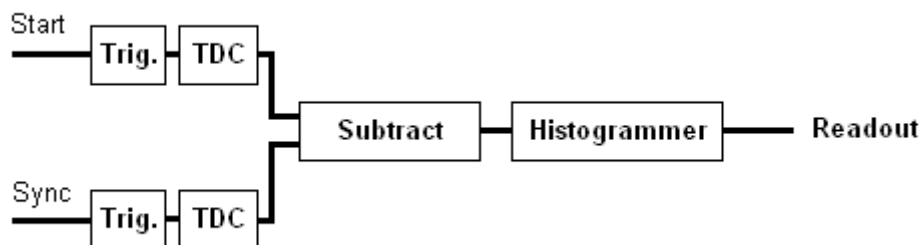
triggering on the zero crossing of the differentiated signal. This has the benefit of adapting to different detector types without a need for changing physical delay lines.

Compared to this, the MultiHarp 150 is different in that it does not use a CFD. The reason behind this is that because of its delay element a CFD requires time to make its “decision” and this time is larger than the MultiHarp’s dead time. Since the short dead time is a precious feature when using high speed detectors, the use of a CFD would spoil the benefit. Indeed HPDs and superconducting nanowire single photon detectors (SNSPDs) have very steep signal edges that do not require a CFD. A simple settable comparator (level trigger) is actually beneficial here. In similarity to the detector signal, the sync signal must be made available to the timing circuitry. Since the sync pulses are usually of well defined amplitude and shape, a level trigger is sufficient to accommodate different sync sources. The MultiHarp 150 therefore uses a level trigger here too.

In historical TCSPC systems the signals from the two input discriminators / triggers are fed to a Time to Amplitude Converter (TAC). This circuit is essentially a highly linear ramp generator that is started by one signal and stopped by the other. The result is a voltage proportional to the time difference between the two signals. In such conventional systems the voltage obtained from the TAC is then fed to an Analog to Digital Converter (ADC) which provides the digital timing value used to address the histogrammer. The ADC must be very fast in order to keep the dead time of the system short. Furthermore it must guarantee a very good linearity (both over the full range as well as differentially). These are criteria difficult to meet simultaneously, particularly with ADCs of high resolution (e.g. 12 bits) as is desirable for TCSPC over many histogram bins.

The histogrammer has to increment each histogram memory cell, whose digital address in the histogram memory it receives from the ADC. This is commonly done by fast digital logic e.g., in the form of Field Programmable Gate Arrays (FPGA) or a microprocessor.

While this section so far outlined the typical structure of conventional TCSPC systems, it is important to note that the design of the MultiHarp 150 is different. Today, it is state-of-the-art that the tasks conventionally performed by TAC and ADC are carried out by a so called Time to Digital Converter (TDC). These circuits allow not only picosecond timing but can also extend the measurable time span to virtually any length by means of digital counters. The MultiHarp 150 uses one such circuit in each input channel and one for the sync input. They independently work on each input signal and provide picosecond arrival times that then can be processed further, with a lot more options than in conventional TCSPC systems. In the case of classic TCSPC, this processing consists of a subtraction of the two time figures and histogramming of the differences. This is identical to the classic start-stop measurements of the conventional TAC approach. The following figure exemplifies this for one detector channel (Start).



The full strength of the MultiHarp design is exploited by collecting the unprocessed independent arrival times as a continuous data stream for more advanced analysis. Details on such advanced analysis can be found in the literature (see section 2.5). In this case the on-board memory is reconfigured as a large data buffer (First In, First Out; FIFO) so that count rate bursts and irregular data transfer are decoupled. This permits uninterrupted continuous data collection with high throughput. This mode of operation is called Time-Tagged Time-Resolved (TTTR) mode or just “time tagging”. Details can be found in section 5.3.

Forward and Reverse Start-Stop Mode

For simplicity it is most convenient to assume that the time delay measurement is directly causal, i.e. the laser pulse causes a photon event and one measures the time delay between laser pulse and the subsequent photon event. However, most conventional TCSPC systems need to give up this logical concept because of the typically high repetition rates of the excitation lasers: Since the time measurement circuit cannot know in advance whether there will be a fluorescence photon, it would have to start a time measurement upon each laser pulse. Considering that conventional TAC conversion times are in the region of .1 to 2 μs , any excitation rate in excess of .5 to 10 MHz would overrun the time measurement circuits. In fact they would most of the time be busy with conversions that never complete, because there is no photon event at all in most cycles. By reversing the start

and stop signals in the time measurement, the conversion rates are only as high as the actual photon rates generated by the fluorescent sample. Historically there were (and had to be) only about 1 to 5 % of the excitation rate and could therefore be handled easily by the TAC / ADC. The consequence of this approach, however, is that the times measured are not those between laser pulse and corresponding photon event, but those between photon event and the next laser pulse (unless a long cable delay is inserted). This still works (by software data reversing) but is inconvenient in two ways:

- 1) Having to reverse the data leads to unpleasant relocation of the data displayed on a true time axis when the time resolution is changed.
- 2) Changing between slow and fast excitation sources requires reconnecting to different inputs.

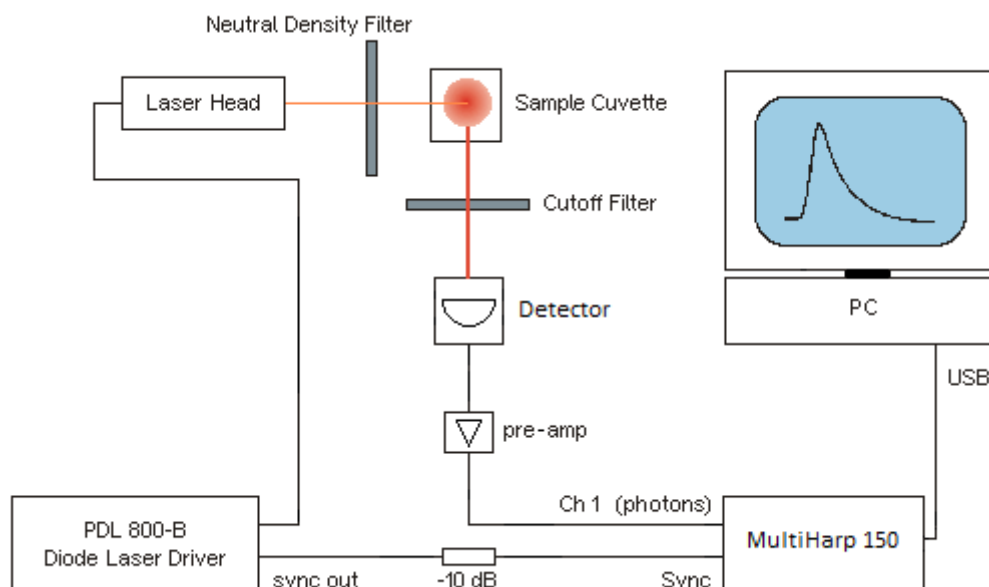
The MultiHarp 150 is very different in this respect, as it allows to work in forward start stop mode, even with fast lasers. This is facilitated by two design features:

- 1) Independent operation of the TDCs of all channels, and
- 2) a programmable divider in front of the sync input.

The divider allows to reduce the input rate so that the period is at least as long as the dead time. Internal logic determines the sync period and re-calculates the sync signals that were divided out. It should be noted that this only works with stable sync sources that provide a constant pulse-to-pulse period. Virtually all currently available fast laser sources meet this requirement within an error band of a few picoseconds. Note: for slow sync sources (< 1 MHz) the sync divider should not be used (set to None). Similarly, the divider must not be used in coincidence correlation measurements (or similar applications) when the sync input receives non-periodic (random) pulses from a photon detector. In summary: The MultiHarp 150 is designed to always work in forward start-stop mode.

Experimental Setup for Fluorescence Decay Measurements with TCSPC

The figure below shows a typical setup for fluorescence lifetime measurements using one input channel of the MultiHarp 150. The picosecond diode laser (PDL 800-B driver with attached laser head) is triggered by its internal oscillator (settable at 2.5, 5, 10, 20 and 40 MHz). The light pulses of typically <70 ps FWHM, are directed toward the sample cuvette via appropriate optics. A neutral density filter can be used to attenuate the light levels if necessary. Upon excitation, the fluorescent sample will emit light at a longer wavelength than that of the excitation light. The fluorescence light is filtered out from scattered excitation light by means of an optical cut-off filter (other configurations may use a monochromator here). Then it is directed to the photon detector, again possibly via some appropriate collection optics, e.g., a microscope objective or a lens.

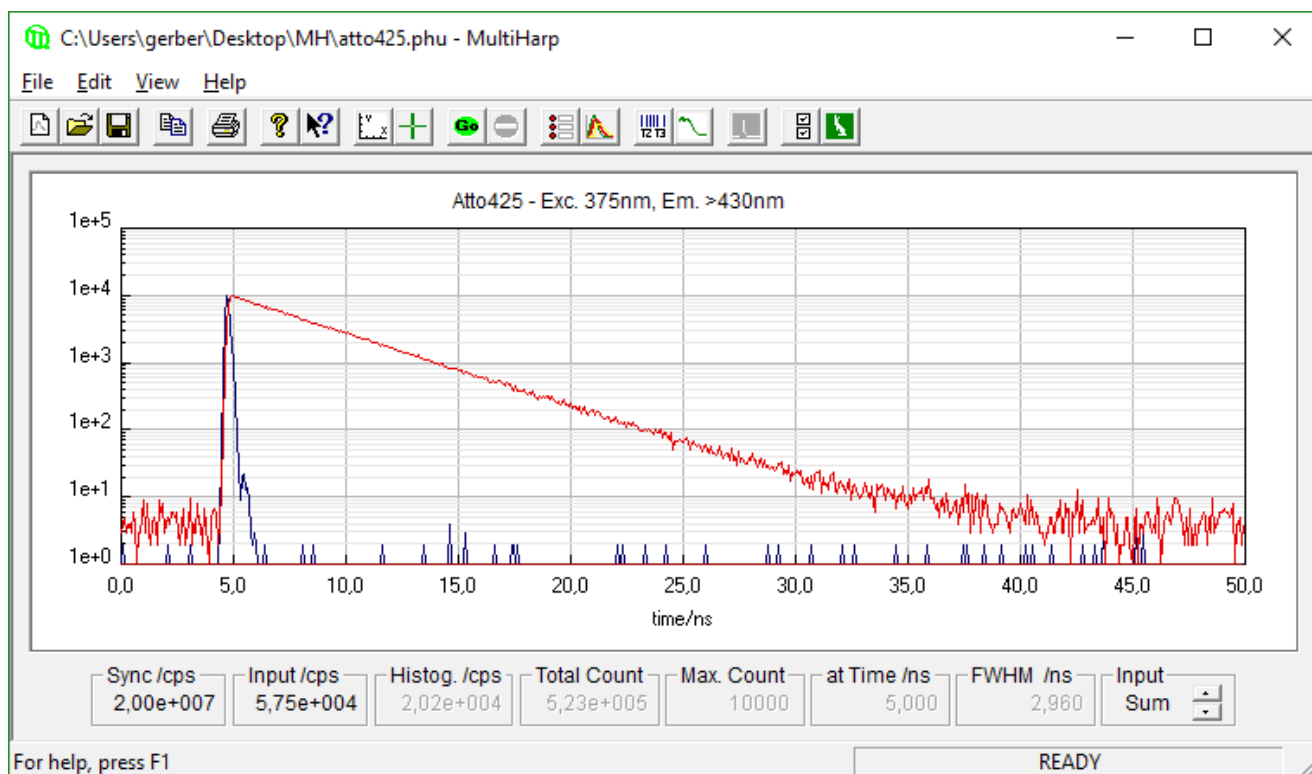


As a photon detector the 10721 PMTs from Hamamatsu are very convenient. They only need a 5 V supply and have an instrument response width of <250 ps, allowing measuring lifetimes even much shorter than this via deconvolution. If a higher time resolution is required, the detector of choice is an MCP-PMT or a HPD. The electrical signal obtained from the detector (small negative pulses of typically -10 to -50 mV) is fed to the TCSPC

electronics via a preamplifier (e.g., PAM 105 from PicoQuant). This gives pulses of -100 to -500 mV. The level trigger is then set for falling edge and e.g., 80 mV. Cabling is double shielded $50\ \Omega$ coax cable. If the detector is a SPAD module with TTL output then the input trigger level must be set to a positive voltage and rising edge. TTL signals must be attenuated (-10 dB) to avoid input damage and to reduce crosstalk. Some SPAD devices like the PDM series from MPD provide negative timing signals that can also be slightly attenuated (-10 dB) for lower crosstalk.

The PDL Series of laser drivers readily provide the electric sync signal needed for the photon arrival time measurement. This signal (a narrow negative pulse) is also fed to the TCSPC electronics via a high quality $50\ \Omega$ coax cable. When using the MultiHarp 150 in combination with the PDL 800-B or a similar PicoQuant laser driver, a 10 dB attenuator can be inserted directly at the sync output of the laser driver. This reduces crosstalk when the detector signals are relatively weak. If the laser does not provide an electrical sync signal (e.g. Ti:Sa lasers), a sync detector (photo diode) such as the TDA 200 must be used.

The following figure shows TCSPC histograms obtained with this kind of setup. Excitation source was a PDL 800-B (PicoQuant) with a 375 nm laser head running at 20 MHz repetition rate. Detector was a PMA Hybrid 06 from PicoQuant with internal amplifier. The cut-off filter was a longpass for >430 nm. The narrower peak (blue curve) represents the system IRF, here dominated by laser and detector. The other curve (red) is the fluorescence decay from a solution of ATTO 425 in water, a fluorescent dye with fairly short fluorescence lifetime (~ 3.7 ns). The count rate was adjusted to $<3\%$ of the laser rate to safely prevent pile-up. The plot in logarithmic scale shows the perfect mono-exponential nature of the decay curve, as one would expect. Note that this is obtained without a deconvolution of the IRF.



The approximate mono-exponential fluorescence lifetime can be obtained from a simple comparison of two points in the curve with counts in the ratio of $1 : 1/e$ (e.g. $100\ 000 : 36\ 788$). For a precise measurement one would perform a numerical exponential fit with IRF deconvolution (typically implemented as an iterative reconvolution). This would result in slightly shorter lifetimes since the IRF broadens the decay. Indeed one can measure lifetimes significantly smaller than the IRF with this method. Additionally, the residues of the fit can then be used to assess the quality of the fit and thereby the reliability of the lifetime measurement. The “EasyTau 2” software package from PicoQuant provides this functionality.

2.5. Further Reading

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12. Bibliography listing all publications with work based on PicoQuant instruments
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13. Product page of the MultiHarp 150
<https://www.picoquant.com/products/category/tcspc-and-time-tagging-modules/multiharp-150-high-throughput-multichannel-event-timer-tcspc-unit>
14. PicoQuant technical and application notes
<http://www.picoquant.com/scientific/technical-and-application-notes>

3. Hardware and Software Installation

3.1. General Installation Notes – Read This First

When handling the MultiHarp 150 system, make sure to avoid electrostatic discharges, especially when connecting cables. Before connecting any signals, carefully study the maximum ratings given in section 8.3.1.

Note that the other PicoQuant TCSPC devices have their own software and their own manuals. The MultiHarp 150 software does not work with these devices and this manual does not relate to them in any meaningful way.

The MultiHarp 150 software version 1.1.0.0 is compatible with Windows 8 and 10¹. Older Windows versions like Windows 7 should still work but are no longer officially supported. Also consider the security risks of using an outdated operating system. Future Windows versions are likely to work but obviously cannot be tested before they are released, and are therefore not formally supported.

In order to use the MultiHarp software with the MultiHarp 150 hardware, a compatible device driver must be installed. The MultiHarp 150 is a 'plug and play' device, meaning that necessary resources and drivers are allocated automatically by the operating system. The software setup will conveniently cater for installing the driver. Note that the current driver does not support Windows 10 with "secure boot" enabled.

On some Windows installations you may need administrator status to perform software setup and de-installation. For the driver installation, it is needed in any case. Installing as Administrator has the benefit that you can install the software for use by all users on that computer even if they have limited access rights. The MultiHarp software will maintain individual settings for each user in the Windows registry. Note that the MultiHarp software does not support running in multiple concurrent user sessions.

The MultiHarp 150 software is also suitable for 64-bit versions of Windows, where a suitable 64-bit device driver will be installed. The MultiHarp software as such is a 32-bit application using WOW64.

3.2. What's New in this Version

MultiHarp 150 software version 1.1.0.0 is the second release for this product, so everything is still relatively new, although many features have been inherited from successful predecessor products. For instance, the file format is the same as for PicoQuant's other TCSPC products and data analysis software such as SymPhoTime 64 and EasyTau 2 is compatible with it from the start. The most significant new feature of the new software version 1.1.0.0 is support for the new 16-channel Model MultiHarp 150 N16. This required a small file format change so that the old software version 1.0.0.0 will not be able to load files from version 1.1.0.0.

3.3. Software Installation

The MultiHarp 150 setup files are normally provided on a CD or DVD supplied with your MultiHarp. If you received the software package by download or any other means of electronic distribution, it will probably be packed in a ZIP-File. In this case you can unzip this file to a temporary hard disk location of your choice and run the software setup from there. On some Windows versions you will need administrator status to perform the setup. For the driver installation you need it in any case.

In order to use the MultiHarp software with the MultiHarp 150 hardware, a device driver must be installed. The MultiHarp 150 is a 'plug and play' device, this means that the necessary resources and drivers are allocated automatically by the operating system. Windows will automatically recognize when such a device is connected and tries to install the appropriate driver. It is recommended to perform the software setup before connecting the device because then Windows will find the driver readily installed. If you skipped that step, Windows may report that it could not install the driver. In that case you can just run the software setup as described below. Alternatively you can direct Windows to search the CD/DVD or, if you downloaded and unpacked the setup files to a hard disk location, direct it to that location. Note that Windows 7 must have the most recent Windows updates in order to install the driver successfully.

To perform the software installation from CD/DVD, insert the installation disk in your CD/DVD drive. Open the CD/DVD drive either from the Windows desktop or with the Windows Explorer. If you downloaded and unpacked the setup files to a hard disk location, open that location. The installer program file containing the complete distribution is named `setup.exe`. Run `setup.exe`. The setup program will guide you through the installation process step by step.



When asked for a destination folder for the new software, please accept the default path or select another according to your program storage policies. This is where the MultiHarp application files will be installed. To avoid confusion, make sure not to specify the path of an older MultiHarp version that you have not uninstalled or that of any other program on your PC.

The default location is: `\Program Files\PicoQuant\MultiHarp150v11`.

Setup will also create a dedicated "program folder" for the new MultiHarp 150 software that will later appear in the Start Menu. You can accept the default folder name or select another according to your own naming policies. However, you should make sure not to specify the folder name of an older MultiHarp version that you have not uninstalled nor the dedicated folder of any other program.

In the chosen destination folder the installer will also create a subdirectory `\filedemo` which contains demo source code for access to MultiHarp 150 data files in various programming languages. Furthermore, another folder `\sampledata` will be created with samples of MultiHarp 150 data files. Other necessary files such as setup information and the device driver will be installed in the standard places in your Windows directory tree.

The setup program will also optionally install the device driver and a *File Info* shell extension that you can use to inspect individual header items of a `*.ptu` or `*.phu` file. This includes the measurement mode. Just right-click on the file in Windows explorer and select *Properties*. Then look at the tab *PQ File Info* and the tab *PQ File Comment*.

After the installation the MultiHarp software should be available in the Windows Start Menu under the custom folder name you chose during setup. If you accepted the default then it will appear under *Programs | PicoQuant – MultiHarp 150 v1.1*.

3.4. Hardware Installation

Make absolutely sure to *avoid electrostatic discharge* when unpacking and handling your MultiHarp 150, especially when connecting cables. Note that PMT detectors operate with high voltage that may discharge through the signal cable. Make sure such detectors are switched off and fully discharged before connecting them.

The MultiHarp 150 has a USB 3.0 super speed interface which is backwards compatible with USB 2.0. In order to obtain the maximum throughput it must be connected through USB 3.0 or 3.1. Consult your PC manual as to whether and where it provides high speed USB 2.0 or super speed USB 3.x connectors. The latter can usually be identified by their blue color. The MultiHarp 150 will not work with a USB 1.x connection. However, all current PCs should provide at least USB 2.0 connectivity. Recent PCs will also provide USB 3.0 or 3.1 out of the box. If the PC has no USB 3.x ports you can install a USB 3.x adaptor card but this may not provide the same throughput as an on-board controller.

Always use a quality USB cable rated for USB 3.0 or 3.1 speed. The cable length must not exceed 5 metres (~16 ft). For best reliability we recommended to use the provided cable of 3 metres length. Note that the USB specification does not allow cable extensions other than dedicated active extension cables or hubs. The MultiHarp 150 should work flawlessly through suitable USB hubs. This is also a valid way of extending the maximum cable length. After a hub, another cable of up to 5 metres is allowed. Note, however, that hubs may lower the data throughput. For the same reasons it is recommended not to connect other devices with high bandwidth requirements to the same hub.

The steps below describe how to connect the signals for your experiment. The inputs for the photon detector(s) and the sync signal are SMA connectors located on the front panel of the MultiHarp 150. They are labelled SYNC and CH1...CHx with x dependent on the model. The sync input is typically used for synchronization with a laser and CH1..CHx are detector signal inputs. The sync input can also be used for detectors if a laser sync is not required, e.g., in antibunching measurements or in generic time tagging applications. All inputs are terminated with 50 Ohms internally. Use quality 50 Ohms coax cables with appropriate connectors. For interfacing to BNC connectors, use standard adapters. Carefully screw on the SMA connectors for sync and detector(s) until they are hand-tight. Do not use wrenches. As of September 2019 your MultiHarp 150 ships with a dedicated SMA connector tool that allows convenient handling in limited space without risk of over-tightening. Note that PMT detectors operate with high voltages that may discharge through the signal cable. Make sure such detectors are switched off and fully discharged before connecting or disconnecting them.

In case of time resolved fluorescence experiments with a pulsed excitation source, the sync signal must be connected to the sync input and the detector signal(s) must be connected to the detector input(s). If coincidence correlation experiments between two (or more) detector signals are to be carried out, you need to decide whether you will be using histogramming or TTTR modes T2/T3. In the case of histogramming and T3 mode connect one detector to the sync input and one or more to the detector inputs. Histogramming and T3 timing will

always be with respect to the sync input. In T2 mode it is possible to determine the relative timing between all inputs but this requires off-line data analysis (see section 5.3).

Connect the signal cable ends to the appropriate signal sources (50 Ω) in your experimental setup. The inputs of the MultiHarp 150 accept positive or negative pulses with peak values of up to +1.2 V or -1.2 V, respectively. The software allows selecting the trigger edge (rising=1 or falling=0) and the trigger level. All inputs should be operated with similar pulse amplitudes to minimize crosstalk. The optimum amplitude range is 100 to 200 mV. Below this range you may pick up noise, above there may be crosstalk. Most PMT and MCP detectors will require a pre-amplifier to reach enough signal level. TTL-SPAD-detectors must be connected through an attenuator or an attenuating inverter (PicoQuant SIA 400). Never connect TTL signals directly, as this may cause damage. Weak PMT detectors should be connected through a 20 dB high speed pre-amplifier. MCP-PMT detectors should be connected through an amplifier with slightly higher gain. When detectors with small signals are being used in combination with laser drivers from PicoQuant's PDL Series, the sync pulses from the laser driver should be attenuated by 10 to 15 dB to fall into the optimum range for smallest crosstalk. Similar attenuation is recommended on the detector signals when detectors with NIM output are used. Suitable attenuator and amplifier devices are available from PicoQuant.

IMPORTANT: switch the high voltage supply of PMTs off and allow their electrodes to discharge before connecting them. Their high voltage charge may damage the pre-amplifier. Observe the allowed maximum ratings for the input signal levels. Above these levels hardware damage will occur. If you are not sure what signals your setup delivers, use a fast oscilloscope to check the signal level and shape before connecting them to the MultiHarp. All signals should have fast rise times of no more than a few ns. Slower signals may degrade timing accuracy.

Do not connect anything other than dedicated hardware to the Sub-D multi-pin connector (control port). It is provided for hardware expansion (notably experiment control) and must not be used otherwise. See section 8.3.2 for pin assignments. It is recommended to start instrument setup without anything connected to the control port.

3.5. Installation Troubleshooting

After completion of the software setup and connecting the MultiHarp the device should be listed in the Windows Device Manager. Right click on the windows start button and select *Device Manager* to check if the device is free of conflicts and / or if the device driver is installed correctly. Under *PicoQuant TCSPC Devices* look for a device named *MultiHarp 150* and inspect its *Properties*.

A common source of problems is the new shutdown behavior of Windows 10. It does not fully shut down by default but goes only into a state similar to hibernation in order to re-start more quickly. When new hardware is installed this can cause problems. If you missed this during hardware installation, do it at least when problems arise. In order to fully shut down Windows hold the shift key while clicking the shutdown button or run the command `shutdown /s /t 0` from the command prompt. It is also possible to permanently configure Windows for proper shutdown via the "power options" dialog.

If the MultiHarp driver is not installed or needs updating you can install it manually. When prompted to search for the driver, direct the driver wizard to search the CD/DVD or, if you downloaded and unpacked the setup files to a hard disk location, direct it to that location. Note that Windows 7 must have the most recent Windows updates applied in order to install the driver successfully.

If things are not working as expected you can also use the Windows system information facilities (*Start > Run > msinfo32*). In the System Information utility inspect *Software environment > System Drivers* to check if the MultiHarp 150 device driver PQCYUSB.SYS is correctly installed.

You can also repeat the software installation if necessary. To do so, first uninstall the software and repeat the setup procedure. Make sure the software is not installed in multiple places. If this does not resolve the problem, try a different computer. If problems persist, see section 7 for support.

3.6. Uninstalling the Software

Before uninstalling the MultiHarp software you should back up all MultiHarp data files you might have created in the MultiHarp installation directory.

Do not manually delete any program files from the installation folder as this will prevent a clean uninstall process.

To uninstall the MultiHarp software from your PC you may need administrator rights (depending on Windows version and security settings). From the list of installed applications select *PicoQuant – MultiHarp 150 vx.x* for un-installation. This will remove all files that were installed by the MultiHarp setup program but not the user data that may have been stored. If there was user data in the program folders or any subfolders, these will not be deleted by the uninstall program. If intended you need to delete these files or folders manually. Nevertheless, it is recommended to back up valuable measurement data before uninstalling the software.

Note that uninstallation of the data acquisition software does not uninstall the device driver. This is because other software may still need it. You can delete the driver software from within Device Manager.

Note also that un-installation of the data acquisition software does not automatically uninstall the PQ File Info shell extension. It can be un-installed as a separate item through the standard Windows Control Panel mechanisms.

4. Software Overview

4.1. Starting the TimeHarp Software

After correct installation the Windows Start Menu or the tiled Windows Start Screen contains a shortcut to the MultiHarp software. To start the MultiHarp software select *PicoQuant – MultiHarp 150 vX.X – MultiHarp 150*. Note that after switching the device on, you need to allow a warm-up period of about 20 minutes before using the instrument for serious measurements. You can use this time for set-up and preliminary measurements.

If the MultiHarp software cannot find a MultiHarp 150 device (or if there are driver problems) it will display a notification message, but it will still start. However, device dependent toolbar buttons and functions of the program will then be disabled. This allows you to use the software without the MultiHarp hardware, e.g., to view or print files on another computer.

It is possible to use up to eight MultiHarp 150 devices on one PC. If multiple devices are installed then the first instance of the software will connect to the first device, the second to the second device and so on. Each of the MultiHarp software instances displays the serial number of the device it uses. An instance that does not find an unused device will open as a file viewer only.

Note, that the various instances of the MultiHarp software will be running completely independent of each other. If your application requires some kind of joint action of multiple MultiHarp 150 devices then you must design your own software based on the MultiHarp 150 programming library MHLib (see separate manual). If your objective is to combine multiple devices to obtain more detector channels you may need to synchronize the clocks of the devices. You also need to consider that it is not possible to prevent Windows from introducing unpredictable delays in communication with the hardware. The latter makes it impossible to, e.g., start measurements on multiple devices at the exact same time. See the MHLib manual and demos for partial solutions.

If the MultiHarp 150 is correctly installed and there are still hardware related errors, you can use the Windows device manager for troubleshooting (see the corresponding section above). If problems cannot be resolved, see section 7 for support. If possible, try the using the device with another computer.

For regular use of the MultiHarp software you may want to create an icon for it on your Windows desktop. You can do this conveniently via a right mouse click on the start menu entry (*send to > Desktop – create shortcut*). Alternatively use the Windows Explorer to locate the file `MultiHarp.exe` in the directory you selected for installation and drag the icon onto the desktop. This will create a link to the MultiHarp executable file.

You can also start up the MultiHarp software directly from a MultiHarp 150 data file by double-clicking on the file or dragging it onto the MultiHarp icon. Note that this may be spoiled if you have other PicoQuant TCSPC devices installed on the same computer. The file name extensions may be assigned to other software.

4.2. The Main Window

The MultiHarp software provides a measurement control interface to the MultiHarp hardware and an online histogram display. Most prominently the MultiHarp main window accommodates the histogram display area.

Above the display area is the toolbar. Here you can access frequently used commands by simple mouse clicks. Next to the toolbar buttons you see the serial number of the device in use. Above the toolbar is the menu bar with additional commands. At the bottom of the histogram display area is a set of 'panel meters' showing count rates, count sums, and histogram peak characteristics. These will be updated continuously, some only when a measurement is in progress. Note the selector at the right of the panel meters. This selects the detector channel the meters are displaying. Instead of a single channel you can also select Sum, which then displays the summed rates from all detector channels. The panel meters can be enlarged by double-clicking them, which is useful when performing optical alignment or similar tasks when the PC monitor is some distance away.

In the top center of the display area a title line is shown. This can be double-clicked to edit the title. When editing the title, note that only the first line will appear in the display. The remaining lines are meant to be used as a file comment. All lines will be stored with the file data, with a maximum of 256 characters.

The main window is resizeable and the actual histogram display will adapt its size accordingly. If you make the window smaller than the minimum histogram display, two scrollbars will permit access to hidden window areas. Note that the actual size of the main window depends on the system font selected. It will scale to the size of the current system font. Screen resolutions below 800x600 are not suitable for serious work with the software.

Note that the position and size of the main window on the screen will be stored in the Windows registry and retrieved upon the next program start. The registry settings are kept separately for each user, provided he / she is logged on with a unique user name. Consult section 6.1 for further main window command descriptions. Toolbar, Menus, panel meters etc. will be explained in the next sections.

At the very bottom of the main window there is a status bar. The leftmost area of the status bar describes actions of menu items as you navigate through menus. Similarly it shows messages that describe the actions of toolbar buttons. The second status bar area from the left shows the current measurement status of the MultiHarp. The rightmost area of the status bar indicates if the <Caps> and <NumLock> keys are latched down.

When the MultiHarp software is running with functional hardware it continuously collects information about the input signals and the current acquisition settings. If these settings together with the input rates indicate possible errors, the software will display a warning icon in the status bar. The warning icon can be clicked to review the list of current warnings together with a brief explanation (see also section 8.1).





















4.3. The Toolbar

The toolbar is displayed across the top of the MultiHarp main window, below the menu bar. The toolbar provides quick mouse access to frequently used commands and tools. Note that some buttons may be grayed out (disabled) depending on the installed software components and / or the state of the hardware.



To hide or display the toolbar, choose Toolbar from the View menu (<Alt>+V T). The following table explains the individual buttons.

| Click... | to... |
|---|---|
|  | open a blank histogram with default control panel settings |
|  | open an existing histogram file. Displays the <i>Open</i> dialog box, in which you can locate and open the desired file. |
|  | save the current histogram data with its current name. If you have not named the file, the <i>Save As...</i> dialog box is displayed. |
|  | copy the currently displayed curves to the clipboard (ASCII export). |
|  | print the currently displayed histogram curves. |
|  | display the <i>About...</i> window. This is where you can determine the version of your MultiHarp software and hardware. Also provides links for updates etc. |
|  | activate context sensitive help. |
|  | launch the axis panel. |
|  | launch the data cursor dialog. |
|  | start measurement based on current MultiHarp control panel settings |
|  | stop measurement and histogram accumulation. |
|  | launch the MultiHarp control panel. |
|  | launch the trace mapping dialog. |
|  | launch the TTTR mode dialog. |
|  | Launch the TTTR mode real-time correlator dialog. |
|  | launch the dialog for monochromator control and TRES setup. |
|  | launch the general settings dialog. |
|  | Launch the White Rabbit dialog |

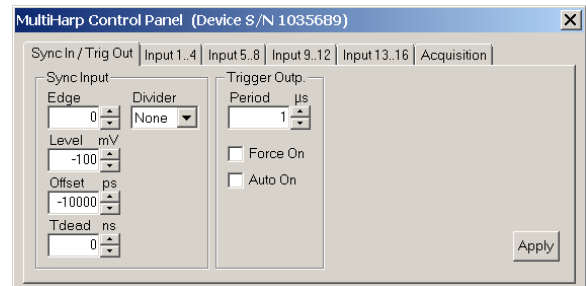
4.4. The Control Panel

The MultiHarp control panel is a dialog box for setting the parameters for hardware adjustment and data acquisition. It is implemented as a 'non-modal' dialog box, i.e. it does not have to be closed before the main window can continue to operate. This way you can make changes to your settings in the control panel and watch their effect on a running measurement in the main window immediately. Nevertheless, you may close the control panel and restore it at any time by clicking the control panel button on the toolbar or pressing <Alt>+C.

The control panel consists of several pages (tabbed sheets) containing groups of edit boxes and other controls for related parameters. These pages and their respective controls are:

Sync-Input and Trigger Output

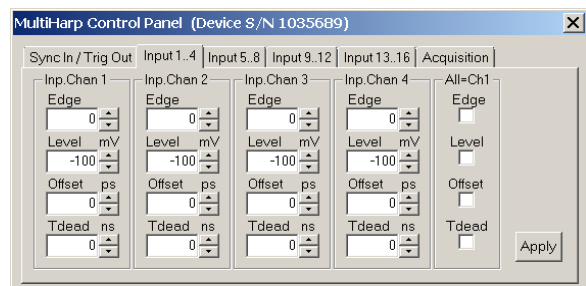
| | |
|----------------------|---------------------------|
| Trigger Edge | edit box and spin control |
| Trigger Level | edit box and spin control |
| Offset | edit box and spin control |
| Tdead | edit box and spin control |
| Sync Divider | edit box and spin control |
| Trigger Outp. Period | edit box and spin control |
| Trigger Op. Force On | tick box |
| Trigger Op. Auto On | tick box |



Inputs 1..4

| | |
|---------------------|---------------------------|
| Zero Cross | edit box and spin control |
| Discriminator Level | edit box and spin control |
| Offset | edit box and spin control |
| Tdead | edit box and spin control |
| All=Ch1, ZeroCr. | tick box |
| All=Ch1, Discr. | tick box |
| All=Ch1, Offset | tick box |

Controls for channels that do not exist remain gray.

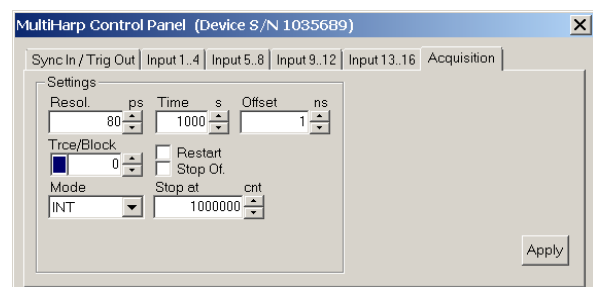


Inputs 5..8, 9..12, and 13..16

These tab pages only appear when the device has more than 4 (or respectively 8) input channels. They have the same controls as the page for inputs 1..4 and follows the same logic of operation.

Acquisition

| | |
|------------------|---|
| Resolution | edit box and spin control |
| Time | edit box and spin control |
| Offset | edit box and spin control |
| Trc/Block | trace colour indicator, edit box and spin control |
| Restart | check box |
| Stop on Overflow | check box |
| Mode | drop down selection box |
| Stop At | edit box and spin control |



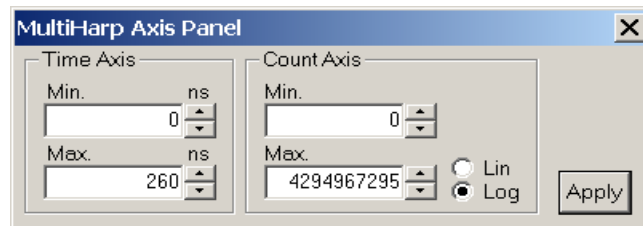
Edit boxes are for keyboard entry. The values must be confirmed with either the <Enter> key or the *Apply* button. The spin controls can be used to increment or decrement the value in the edit box. In this case the changes take effect immediately without need for hitting <Enter> or clicking *Apply*. Check boxes have their denoted effect when the tick is shown. They can be toggled with a mouse click. Groups of radio buttons are like check boxes but mutually exclusive.

Note that the settings of the control panel as well as the positions of control panel and main window on the screen will be stored in the Windows registry and retrieved during the next program start. The registry settings are stored separately per user. When a MultiHarp data file is loaded, the control panel settings will change to reflect the settings stored in that file.

The individual control panel items are discussed in section 6.4 in the Controls and Commands Reference.

4.5. The Axis Panel

The axis settings panel is a dialog box for setting the axis range for the histogram display in the main window. It is implemented as a 'non-modal' dialog box, i.e. it does not have to be closed before the main window can continue its operation. This way you can make changes in the axis panel and watch their effect in the main window immediately. Nevertheless, you may close the axis panel and restore it at any time by clicking the axis panel button on the toolbar or pressing <Alt>+A. The panel will also open if you double-click the axes in the main window.



The axis panel consists of two groups containing edit boxes and other controls for related parameters. These groups and their respective controls are:

Time Axis

| | |
|----------------|---------------------------|
| <i>Minimum</i> | edit box and spin control |
| <i>Maximum</i> | edit box and spin control |

Count Axis

| | |
|--------------------|---------------------------|
| <i>Minimum</i> | edit box and spin control |
| <i>Maximum</i> | edit box and spin control |
| <i>Linear</i> | radio button |
| <i>Logarithmic</i> | radio button |

The edit boxes are for keyboard entry. The values must be confirmed with either the <Enter>-key or the *Apply*-button. The spin controls can be used to increment or decrement the values in the edit box. In this case the changes take effect immediately without need for hitting <Enter> or clicking *Apply*. The mouse wheel can be used for fast spins as long as the cursor is in the corresponding edit box. Check boxes have their denoted effect when the tick is shown. They can be toggled with a mouse click. Groups of radio buttons are like check boxes but mutually exclusive.

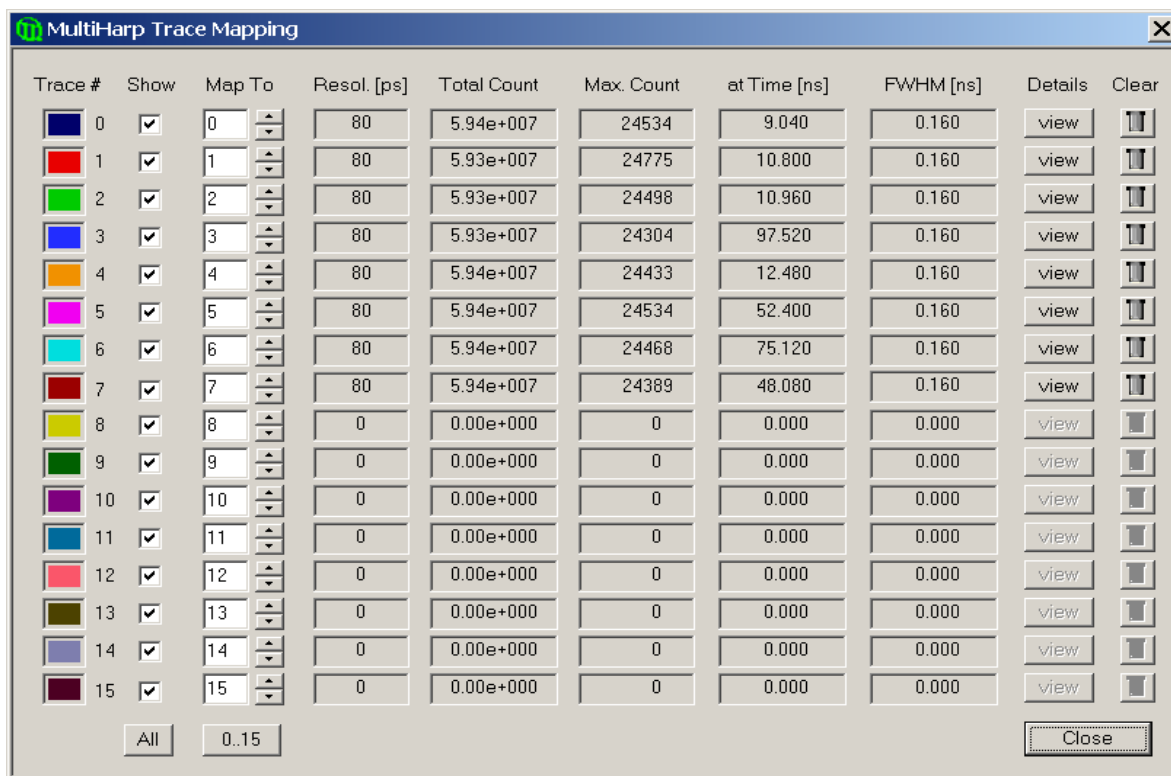
Note that the settings of the axis panel as well as the positions of the panel on the screen will be stored in the Windows registry and retrieved during the next program start. The registry settings are stored on a per user basis.

The individual axis panel items are discussed in section 6.5. "Axis Panel" in the Controls and Commands Reference.

Hint: to begin, use logarithmic display to make sure even weak signals can be seen.

4.6. The Trace Mapping Dialog

The MultiHarp software can record and store histograms in up to 512 memory blocks. Out of these up to 16 curves can be displayed at the same time. The *Trace Mapping* dialog is used to select the curves to display. It also allows to view curve details and cleanup. You can use the *Trace Map* button on the Toolbar or click the curve colour indicator in the control panel to launch the dialog.



In the *Trace Mapping* dialog you can tick the individual boxes 'Show' to display a curve. You can also select the index of the memory block you wish to map the individual display curves to.

The dialog also provides some statistics on each curve (central matrix of figures). *Resolution* is the bin width of the histogram in picoseconds. Next is the *Max. Count*, the count in the highest point of the curve. The column *at Time* shows the time corresponding to the *Max. Count* bin. Leftmost there is the Full Width Half Maximum (*FWHM*) of the curve peak (usually meaningful only for IRF traces).

There are also some buttons for frequently required actions: The button 'view' can be clicked to see more detailed curve information such as time of recording, acquisition settings and count rates. The button *All* can be clicked to tick all traces as shown. The button *0..15* can be clicked to set the default mapping of trace 0..15 to block 0..15. The *Clear* buttons (trash cans) can be clicked to delete the contents of individual blocks.

Note that the Trace Mapping dialog is non-modal. This means the dialog can remain open while a measurement is in progress, so that adjustments can be made under immediate visual control, similar to the operation of the control panel. Note also that a measurement can be running in a block that is not mapped or shown.

The individual trace mapping panel items are discussed in the section 6.6. "Trace Mapping" in the Controls and Commands Reference.

4.7. Other Dialogs

In order to keep this manual readable, the dialogs described here are limited to the most important ones the reader should know about before starting practical work with the software. Additional dialogs will be described implicitly in the following sections in the context of specific measurement tasks. For information on all other dialogs the user is kindly referred to the controls and commands reference (section 6) or consult the on-line help facility of the software. Pressing F1 in any given dialog will open the corresponding help page.

5. Specific Measurement Tasks

5.1. Setting Up the Input Channels

This section provides help and instructions for the first basic steps of setting up the instrument. However, if you are running TCSPC measurements for the first time we strongly recommend you read the primer on TCSPC in section 2 first. Also consider the literature listed there.

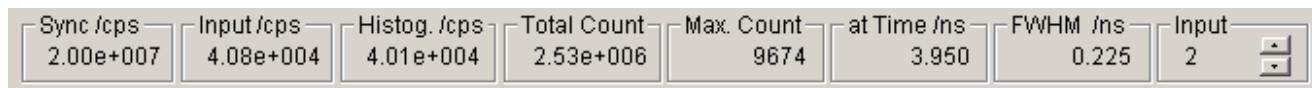
In order to acquire any data, the input channels and the sync input of the MultiHarp 150 must be set to match their electrical input signals. The MultiHarp input channels are designed identically. All inputs have a programmable level trigger (comparator) allowing the selection of the trigger edge (rising=1, falling=0) and the trigger level in mV. For specification details see section 8.3.1 and take note of the maximum ratings.

In case of coincidence correlation experiments using two or more detectors, the input channels are typically used for one detector each and data is collected in T2 mode. In that case the sync input can (but need not) be used for a detector as well. If a quick visualization of coincidence is required, it is also possible to use histogramming mode. In that case, one detector is connected to the sync input. Coincidence histograms will be collected for each input channel with respect to the sync input. In time-resolved fluorescence measurements with a pulsed excitation source (typically a laser) the sync input receives a sync signal from the laser. Here we focus on the latter, more common case. Perform the following steps to set up the input discriminators.

Using the Panel Meters

At the bottom of the main window you find a set of panel meters. These are very important during set-up. The meters showing units of cps (counts per second) in their title are rate meters. The leftmost rate meter shows the sync input rate. The next meter shows the channel input rate. Note the selector at the far right of the panel meters. This selects the channel the rate meters (except sync) are referring to. Select the channel you are currently setting up. Note that the rate meters use a fixed gate time of 100 ms. Their accuracy at low rates is therefore limited. They really only serve as a quick means of diagnostics and should not be used to obtain definitive measurement results.

The other meters show the histogramming rate, the total count in the histogram, the maximum (peak) count and the position of the maximum. All of these values depend on the input selector.



Note that the Input selector also has a selection option labelled *Sum*. In this case the meters are fed from the sum of all input channels. This is the default.

Setting up the Sync Input

For typical fluorescence decay measurements, the MultiHarp needs an electrical sync signal from the light source. The PDL Series of diode lasers from PicoQuant provides this signal directly. If the laser does not provide an appropriate electrical sync signal (e.g., Ti:Sa lasers), a sync detector (photo diode) such as the TDA 200 must be used. The sync signal must consist of pulses with steepness and amplitude matching the specifications of the MultiHarp 150 and the trigger edge must be set to the leading edge (rising=1, falling=0). For example, a NIM type signal is appropriate. This is a steep negative pulse (0.5 to 10 ns wide, active edge falling) of typically -800 mV into 50 Ω . The MultiHarp 150 can actually handle ± 1.2 V but large amplitudes may cause excess interference and crosstalk between the inputs. Amplitudes around 100 to 200 mV (on all inputs) are best in terms of timing accuracy and lowest histogram ripple due to crosstalk. It may therefore be advantageous to attenuate NIM pulses by 10 or 15 dB. As a rule of thumb: lowest crosstalk is achieved by using signals of similar amplitude on all inputs. SMA in-line attenuators of suitable bandwidth can be used to adjust this. The trigger level is adjustable for optimum timing accuracy. Initially you should set it to half of the sync pulse amplitude. Later it can be fine-tuned empirically.

Unless you are sure what kind of signal your sync source delivers, use a fast oscilloscope (50 Ω input) to check the pulse shape, polarity and amplitude. The leading edge should be steep (ideally 2 ns rise/fall time or faster), there should also be no excessive ringing. The pulse width should be at least 0.4 ns, the upper limit is not critical.

If the signal is satisfactory, connect the source to the sync input and start the MultiHarp software if it is not yet running. A detector signal (CH1..CHx) is not required at this point but it does no harm if it is also connected. Open the MultiHarp control panel. Leftmost, there is a group of controls for the sync input. Find the edit box and spin control for the trigger level. The level should be set to a value around half of the amplitude of the sync pulses. Next, set the appropriate signal edge (typically the leading edge of your signal). The code 0 means falling, 1 means rising. Then look at the sync divider. It must be set so that the sync rate divided by the shown divider value remains under 78 MHz, e.g., for a laser with 80 MHz repetition rate it must be set to 2. For a slow source such as a flash lamp it must be set to None. Tdead is a programmable dead-time for suppression of afterpulsing artefacts of some detectors. On the sync input it is rarely needed. Leave it at 0 for now.

If the sync source is active, the sync rate will now be displayed at the bottom left rate meter in the MultiHarp main window. Note that the rate meter internally corrects for the chosen divider setting, so that the meter always shows the undivided input rate. The meter display should therefore match the rate delivered from the source. The rate meter will be refreshed every 0.1 to 1 seconds, as determined by the value in the General Settings Dialog. The sync rate should be displayed very accurately unless it is very low. Large fluctuations or occasional zeros indicate an incorrect discriminator level setting or an unstable sync signal. Try varying the discriminator level to obtain a stable sync rate display. If the rate is stable and at the expected value, you can proceed to setting up the other inputs. A last fine tuning of the trigger level can be done when the detector inputs are up and running. Note that it may be impossible to get a stable sync rate reading at very low sync rates. This is because the rate counter uses a prescaler and a gate time of 100 ms and if the signal period is too low the readings will fluctuate. You will then have to set the level based on what you know about the pulse amplitude and verify it in an actual measurement.

Setting up the Photon Detector Inputs (Channel 1...N)

Depending on the model, your MultiHarp 150 may have 4, 8 or 16 input channels. As noted before, the MultiHarp 150 input channels are electrically identical by design. The input signals must consist of pulses with steepness and amplitude matching the specifications and the trigger edge should be set to the leading edge (rising=1, falling=0). For example, a NIM type signal is appropriate. This is a steep negative pulse (0.5 to 10 ns wide, active edge falling) of typically -800 mV into 50 Ω . The MultiHarp 150 can actually handle ± 1.2 V but large amplitudes may cause excess interference and crosstalk between the inputs. Amplitudes around 100 to 200 mV (on all inputs) are best in terms of timing accuracy and lowest histogram ripple. It may therefore be advantageous to attenuate NIM pulses by 10 or 15 dB. As a rule of thumb: Lowest crosstalk is achieved by using signals of similar amplitude on all inputs. SMA in-line attenuators of suitable bandwidth can be used to adjust this. Note that popular TTL-SPAD-detectors (e.g., Perkin-Elmer/Excelitas SPCM-AQR) deliver positive pulses of ~3 V and must be connected through an attenuator or a pulse inverter with attenuation (PicoQuant SIA 400). Connecting TTL signals directly will cause damage to the MultiHarp 150!

PMTs should be connected through a preamplifier (10 to 20 dB). MCP-PMT detectors should be connected through an amplifier with slightly higher gain. All accessories are available from PicoQuant. Be sure to switch the high voltage supply of PMTs off and allow their electrodes to discharge before connecting / disconnecting them. Their high voltage charge may damage the preamplifier. Observe the allowed input signal levels including those of the pre-amplifier. Again, in a new experimental setup, to be absolutely sure, please check your detector pulses as well as the preamp output with a fast oscilloscope. Start timing is on the leading edge, so it should be steep. Ringing and overshoot should be as small as possible. Do not over-illuminate the detector to avoid damaging it.

If the signals are appropriate, connect a detector at channel 1. Starting with only one detector makes the first steps easier. Preliminary adjustments can actually be done with uncorrelated light, e.g. daylight. To protect your detector, use strongly attenuated light, even when the detector is off. Start the MultiHarp software and open the Control Panel. Look at the Control Panel section for the chosen channel. Initially set the trigger level to half the expected pulse height and set the trigger edge (rising=1, falling=0) to match the leading edge of your detector pulses. Tdead is a programmable dead-time for suppression of afterpulsing artefacts of some detectors. Leave it at 0 for now.

Using the input selector at the far right of the rate meters, select the channel you are currently using. The rate meter next to the sync rate meter will now show the input rate at that channel. Make sure that there is actually a signal coming in (some strongly attenuated light on the detector) and try to adjust the trigger level. To monitor this, watch the count rate meter. Precise tuning of the level settings is only critical for PMTs or HPDs. For such detectors an initial setting of the trigger level of -50 mV (assuming a non-inverting amplifier, otherwise +50mV) should be approximately right to suppress electrical noise. Moving this level towards zero should result in an increased count rate (probably mostly noise). If you move the trigger level farther away from zero, you will see a reduced rate that will incur losses of true counts if you go too far. Ideally you should stay just slightly above the noise level. If you still see millions of counts per second at -100 mV, reduce the detector illumination. Your de-

detector may be at risk at such high count rates. You can then also try out responsivity to illumination changes. For SPAD detectors that typically deliver pre-shaped pulses of constant amplitude the setting of the trigger level is very simple. Just set the trigger level to approximately half of the pulse amplitude.

To actually collect histograms, select a measurement range large enough (determined by the chosen resolution) and a display range to cover your sync interval (i.e. $1/f_{\text{sync}}$) if possible. Set Offset = 0 and StopAt = 4,294,967,295. Start a measurement in oscilloscope mode with e.g., 1 second acquisition time (see the next section for instructions). Once counts are coming in, you can try to set an upper limit for the count axis so that the histogram is scaled for best viewing. Start with logarithmic display so that weak signals can be seen.

If you used uncorrelated light so far, you may now want to move to time-correlated measurements. With a detector signal that is e.g., induced by a laser or a 'fake' electrically derived from the laser sync, you can try to obtain a histogram that should be a narrow peak, as opposed to the flat distribution of uncorrelated light. This requires an experimental setup that delivers the signals at the two input channels with a more or less constant relative delay. In histogramming mode this delay must be such that the Channel 1..N (detector) signal comes later than the sync signal (forward start-stop mode). In addition, the delay must be chosen so that it fits in the measurement range of the histogrammer. To obtain such a timing it may be necessary to adjust the input offsets or the relative cable lengths, while also considering any optical delays.

If the rough setup is complete and a time-correlated signal peak is present in the histogram you may want to switch to a higher resolution. For optimization purposes you can then try to slightly vary the trigger levels (including sync) for best timing response. The Full Width at Half Maximum (FWHM) of the signal peak is displayed on-line as a figure of merit for instrument response. If there is no clear optimum for any level, return to the center of its stable working range. You can then also adjust the input offsets to place the signal properly within the time axis boundaries of the sync frame. You can also adjust your axis limits to optimize the display. Note that you can use the mouse wheel for quick changes of any spin control. Note also that you can double-click all the rate meters to enlarge them as separate windows. This is useful for optical alignment work if the PC screen is some distance away from your optical table.

Input Troubleshooting

Whenever there is a problem, first check your cabling, detector power supply, and sync source. To be absolutely sure, check the signals with a fast oscilloscope (50 Ω input!). Never use TTL signals directly. Never try to deliver the signals to multiple 50 Ω loads in parallel, using simple T-pads. If you have multiple detectors, try to test your setup with one detector first to keep things simple. Never forget to let detectors using high voltage discharge before connecting/disconnecting.

If you cannot get a stable sync rate reading with expected values, there may be several reasons:

- there is no proper sync signal (voltage, polarity, pulse width, frequency)
- the sync trigger level setting is inappropriate
- the sync divider setting is inappropriate
- Note that at small rates the meter display may fluctuate between some discrete values.

Reasons for zero input channel counts are:

- wrong input selection for the rate meters
- no or inappropriate signal (voltage, polarity, pulse width, frequency)
- inappropriate trigger levels
- preamplifier or detector failure

Note that all meters are updated at the display refresh rate you have selected in the general settings dialog. In oscilloscope mode the update frequency is equal to the acquisition time. If nothing seems to happen at all, you may be in oscilloscope mode with a very long acquisition time set.

Once count rates > 0 are being displayed you should then also see counts appearing in the histogramming rate meter, if your measurement range is large enough. Make sure the offset is set to 0. If there are histogramming counts but no histogram is building up on the screen, check the time and count axis bounds. It is best to start with a wide display range and then narrow it down. Similarly, a logarithmic count axis setting is the safest way to

see even small histograms. If your measurements stop earlier than expected, make sure the Stop At level is not set to less than 4,294,967,295 unless you have experimental reason to limit the counts.

During the set-up process you should pay attention to the warning icon that may appear at the bottom right of the main window. When the MultiHarp software is running with functional hardware it continuously collects information about the input signals and the current acquisition settings. If these settings in combination with the input rates indicate possible errors, the software will activate the warning icon.



The warning icon can be clicked to display a list of current warnings together with a brief explanation of each warning (see also section 8.1). Note that the software can detect only a small subset of possible error conditions. It is therefore not safe to assume “all is right” just by seeing no warning. On the other hand, if any of the warnings turns out to be an unnecessary nuisance, e.g., because your specific measurement conditions will expectedly cause it, you can disable that warning via the general settings dialog (see section 6.7).

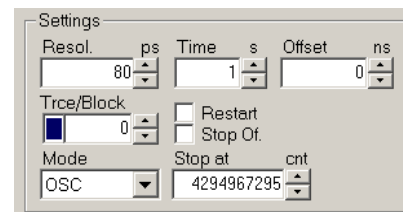
5.2. Setting Up and Running Interactive Measurements

The primary mode of operation of the MultiHarp software is interactive histogramming. This is what the main window of the software is dedicated to. The user can set up measurement parameters, start measurements and immediately see histogram data on the screen. In further sections, e.g., on TTTR mode, you will learn about other modes of operation with less user interaction that will collect data straight to disk without immediate visualization. Here, we focus on the interactive histogramming mode of operation.

To set up measurement parameters use the MultiHarp control panel. The control panel can be opened by clicking the control panel button on the toolbar or by pressing <Alt>+C.



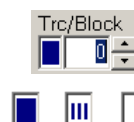
In the control panel section 'Acquisition - Settings' you can set the resolution (time per bin), the offset, the measurement time, and the block of memory to use for this measurement. To begin, use a measurement time of 1 second and an offset of 0. There are always 65,536 time bins per histogram. Histograms can be recorded and stored in 512 memory blocks. Out of these up to 16 curves can be displayed and one 'active' block can be used for a measurement. You can designate the active memory block you wish to use for the next measurement by selecting the block number in the control panel. Never forget to select a new block when collecting new data and old data is intended to be preserved.



Use the trace mapping dialog to select up to 16 curves for display. Make sure the memory block you measure into is mapped to a display trace that is switched on, so that you can see the trace. You can reach the trace mapping dialog from the toolbar or by clicking on the trace colour indicator next to the block selector.



The color indicator shows the trace color that the chosen block is currently mapped to. If it is a solid square, the curve is mapped and shown. If it is mapped but not shown, the indicator shows a small striped square. If the curve is not even mapped for display the indicator remains white.



There are two basic histogramming modes for interactive measurements: Oscilloscope and Integration mode. Oscilloscope mode repeatedly collects histograms with a fixed measurement time and displays them on the screen. This lets you see fast changes in the histogram, e.g., for optical setup and adjustments. Usually this only makes sense with relatively strong signals and short acquisition times. Integration mode is usually operated with longer acquisition times. In this case the histogram continues to grow over a longer time and the display is updated at regular intervals, so that the accumulation process can be observed.



To start a measurement with the current control panel settings, use the start button (GO) on the toolbar or press <Alt>+G.



To stop a measurement use the stop button on the toolbar or press <Alt>+S.



Note that a measurement may automatically stop and / or restart, dependent on the current settings of 'stop on overflow' and 'restart' in the control panel.

Note: to actually run a meaningful measurement you will need to set up the input channels, most importantly with appropriate voltage levels as outlined in the previous section. Also allow a warming-up period of about 20 minutes (depending on lab temperature) before using the MultiHarp for final measurements. You can use this time for set-up checks and preliminary measurements.

The rate meters (bottom of main window) permit visual control of the data acquisition. Note the selector at the far right of the rate meters. This selects the channel the rate meters are referring to. Select the channel you are currently using. The meters are often too small to view from further away, e.g., when adjusting the optical setup. You can in this case simply double-click the rate meter of interest. This opens a large meter display that you can then re-size and move on the screen as you like.

Once you have established standard settings for your experimental setup you may want to save them to a file. The control panel settings can then be recalled at any time by loading that file. The program stores all settings together with the histogram data of those curves in memory (max. 512) that have been filled by a measurement. In addition to this, all settings are stored in the Windows registry, so that at program startup you find the control panel as it was when you last closed the MultiHarp software.

5.3. Time Tagged Mode Measurements

Time–Tagged Time–Resolved (TTTR) mode allows the recording of individual count events directly to hard disk without immediately forming histograms.

In classic TTTR mode, in addition to the start–stop timing with picosecond resolution, the timing of the events with respect to the beginning of the whole measurement is recorded in the event records. This is particularly interesting where the dynamics in a fluorescence process are to be investigated. The availability of the time–tags permits photon burst identification, which is of great value e.g., for Single Molecule Detection (SMD) in a liquid flow. Other typical applications are Fluorescence Correlation Spectroscopy (FCS) and Burst Integrated Fluorescence Lifetime (BIFL) measurements. Together with an appropriate scan controller, TTTR mode is also suitable for ultra fast Fluorescence Lifetime Imaging (FLIM).

The MultiHarp 150 actually supports two different Time–Tagging modes, T2 and T3 mode, which will be explained further below. When referring to both modes together we use the general term TTTR.

5.3.1. System Requirements

In cases where the Time–Tagging modes are to be used with high continuous count rates (say > 5 Mcps) the PC system must meet some special performance criteria. The reason for this is the relatively large amount of data being generated in TTTR mode. In order to prevent an overflow in the recording, the data must be transferred to the computer in real–time. This requires a modern PC with a fast I/O subsystem. A recent, at least dual core CPU running at 2 GHz or more is required. For the best possible performance in TTTR mode a modern hard disk with high throughput is recommended. If it is intended to make use of the full TTTR throughput of a MultiHarp (up to 90 Mcps) then the hard disk must be able to handle sustained write rates of 360 MBytes/s. This can be achieved with RAID arrays or modern solid state disks. Network storage is usually too slow.

5.3.2. T2 Mode

In T2 mode all timing inputs of the MultiHarp 150 including the sync input are functionally identical. There is no dedication of the sync input channel to a sync signal from a laser. It may be left unconnected or can be used for an additional detector signal. In this case the sync divider must be set to “None”. Usually the regular inputs CH1..N are used to connect photon detectors. The events from all channels are recorded independently and treated equally. In each case an event record is generated that contains information about the channel it came from along with the arrival time of the event with respect to the overall measurement start. The timing is recorded with the highest resolution the hardware supports (80 ps). Each T2 mode event record consists of 32 bits. There are 6 bits for the channel number and 25 bits for the time–tag. If the time tag overflows, a special overflow record is inserted in the data stream, so that upon processing of the data stream a theoretically infinite time span can be recovered at full resolution. Dead times exist only within each channel but not across the channels. Therefore, cross correlations can be calculated down to zero lag time. This allows for powerful applications such as coincidence correlation and FCS with lag times from picoseconds to hours. Autocorrelations can also be calculated at the full resolution but of course only starting from lag times larger than the dead time.

The 32-bit event records need to be continuously streamed to the host PC, ideally without losses. Given the dead-time of the TDCs of $T_d=650$ ps it is theoretically possible that event records are generated at a maximum rate of $1/T_d = 1,538$ GHz. The total rate increases even more when multiple channels are used. It is obvious that such data rates cannot be transferred over USB continuously without losses. On the other hand, given typical photon statistics one can (and should) distinguish between peak rates and average rates. The latter are typically much smaller than the peak rates during bursts. The system is therefore designed to use FIFO (First In First Out) buffers that can temporarily hold a certain number of events in a burst so that the bus transfer must only deal with moderate average rates. In order to handle the maximum input burst rate the MultiHarp 150 uses in each channel a fast front-end FIFO buffer that can handle bursts of up to 2000 events at the maximum rate of $1/T_d = 1,538$ GHz. This is followed by a large but slower FIFO buffer for all channels, capable of holding up to 134,217,728 event records. This large secondary FIFO buffer ensures that no data is lost due to inevitable task switches and interruptions of the USB transfers on the host PC side. Even if the average read rate of the host PC is limited, bursts with much higher rate can be recorded for some time. Only if the average count rate exceeds the readout speed of the PC over a long period of time, a FIFO buffer overrun can occur. In case of a FIFO buffer overrun the measurement must be aborted because data integrity cannot be maintained. However, on a modern and well configured PC a sustained average count rate of up to 90 Mcps is possible. This total transfer rate must be shared by the inputs used. For all practically relevant photon detection applications the effective rate per channel is more than sufficient. If there are very intense bursts at the input of the front-end FIFO it may happen that events are lost. This is indicated to the software by means of a hardware flag so that the

user can be informed of such losses. The user must then decide if the losses can be tolerated for the given experiment (see also section 8.1 on warnings).

For maximum throughput, T2 mode data streams are normally written directly to disk, without preview other than count rate and progress display. However, it is also possible to analyze incoming data "on the fly". The MultiHarp software provides a real-time correlator for preview during a T2 mode measurement (see section 5.3.7). Other types of real-time processing must be implemented by custom software. The MultiHarp software installation provides demo programs to show how T2 mode files can be read by custom software (see the folder `file-demo` under the chosen software installation folder). The implementation of custom measurement programs requires the MultiHarp programming library, which is provided as a separate software package on the distribution media or as download. Alternatives for advanced T2 data collection and analysis are the SymPhoTime and QuCoa software suites offered by PicoQuant. SymPhoTime is focused on typical life science applications while QuCoa is oriented towards typical quantum optics applications.

5.3.3. T3 Mode

In T3 mode the sync input is dedicated to a periodic sync signal, typically from a laser. As far as the experimental setup is concerned, this is similar to classic TCSPC histogramming. The main objective is to allow for high sync rates which could not be handled in T2 mode. Accommodating the high sync rates in T3 mode is achieved as follows: First, the sync divider is employed as in histogramming mode. This reduces the sync rate so that the channel dead time is no longer a problem. The remaining problem is now that even with the divider, the sync event rate may still be too high for collecting all individual sync events like in ordinary T2 mode. Considering that sync events are not of primary interest, the solution is to record them only if they arrive in the context of a photon event on any of the input channels. The event record is then composed of two timing figures: 1) the start-stop timing difference between the photon event and the last sync event, and 2) the arrival time of the event pair on the overall experiment time scale (the time tag). The latter is obtained by simply counting sync pulses. From the T3 mode event records it is therefore possible to precisely determine which sync period a photon event belongs to. Since the sync period is also known precisely, this furthermore allows reconstructing the arrival time of the photon with respect to the overall experiment time.

Each T3 mode event record consists of 32 bits. There are 6 bits for the channel number, 15 bits for the start-stop time and 10 bits for the sync counter. If the counter overflows, a special overflow record is inserted in the data stream, so that upon processing of the data stream a theoretically infinite time span can be recovered. The 15 bits for the start-stop time difference cover a time span of $32,768 \times R$ where R is the chosen resolution. At the best possible resolution of 80 ps this results in a span of 2621 ns. If the time difference between a photon and the last sync event is larger, the photon event cannot be recorded. This is the same as in histogramming mode, where the number of bins is larger but also finite. However, by choosing a suitable sync rate and a compatible resolution R , it should be possible to reasonably accommodate all relevant experiment scenarios. R can be chosen in a wide range, starting with the card's base resolution and then continuing by repeatedly doubling of the time bin width.

Dead time in T3 mode is the same as in the other modes (hardware model dependent). Within each photon channel, autocorrelations can be calculated meaningfully only starting from lag times larger than the dead time. Across channels dead time does not affect the correlation so that meaningful results can be obtained at the chosen resolution, all the way down to zero lag time. This requires custom software.


The 32 bit event records are queued and forwarded to the host PC in the same staggered FIFO architecture as described in the section on T2 mode above. Accordingly, a sustained average count rate of up to 90 Mcps is possible in T3 mode too, while now the sync events do not consume any transfer bandwidth.

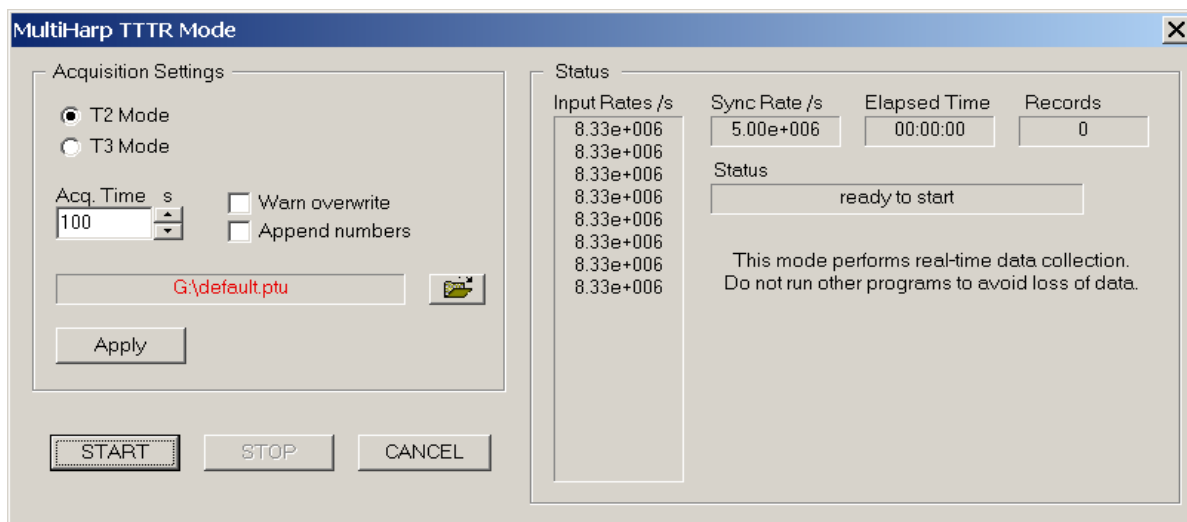
For maximum throughput, T3 mode data streams are normally written directly to disk. However, it is also possible to analyze incoming data "on the fly". One such analysis method is the on-line correlation implemented in the MultiHarp software. Other specialized analysis methods must be implemented via custom software. The MultiHarp software installation provides demo programs showing how T3 mode files can be read (see the folder `filedemo` under the chosen software installation folder). The implementation of custom measurement programs requires the MultiHarp programming library, which is provided as a separate software package on the distribution media or as download. An alternative for advanced T3 mode data collection and analysis is the SymPhoTime software suite offered by PicoQuant.

5.3.4. Running a basic TTTR Mode Measurement

A TTTR mode measurement (T2 or T3 mode) will typically be started after all control panel settings have been tested in normal interactive histogramming mode (oscilloscope or integration). The acquisition time (measurement time) and the file for saving the data are the only parameters that can be set separately.

A typical approach to set up a TTTR mode measurement would be by first starting oscilloscope mode with an acquisition time of e.g., 1 second. Then all control panel settings should be optimized to reliably obtain the expected data.

Once all settings are satisfactory, click the “TTTR Mode” button  on the toolbar. This will bring up the “TTTR Mode” dialog.



The dialog section *Acquisition Settings* allows selecting the measurement mode (T2 / T3), overall acquisition time, and file name. Note that switching between T2 and T3 mode takes some time because the hardware must be reconfigured. Normally such a switching should not occur often because the two modes usually require a different experiment setup.

The section *Acquisition Settings* also has two tick boxes for the handling of existing files. You can turn on a warning and / or automatically have numbers appended to the file names, so that you can conveniently perform series of measurements. The file name is shown in red if the file already exists. The button with the file icon will open a standard Windows file dialog. You can select an existing file or choose a new name. The MultiHarp TTTR mode files have the extension ".ptu". For maximum count rate throughput you should choose a file destination on a fast local hard disk as outlined above. Network drives are often too slow.

The dialog section *Status* shows elapsed time, the count rates and the number of collected records. Below these figures is a status line showing what is currently happening. Further below there are buttons for Start, Stop and Cancel. Start and Stop control the actual TTTR measurement run. Cancel is for leaving the TTTR mode dialog.

The TTTR mode measurement will start as soon as you click the Start button. You will then be able to watch the progress of your measurement in the status boxes showing the elapsed measurement time and the number of events that have been recorded. Note that this includes overflow and marker records. Therefore, the number of records will be somewhat larger than the true photon counts. The overflow and marker records will later be filtered out by the data analysis / processing software.

Next to the status line you may see a warning icon. When the TTTR mode dialog is running it continuously collects information about the input signals and the current acquisition settings. If these settings in conjunction with the input rates indicate possible errors, the software will display the warning icon. The warning icon can be clicked to show a list of current warnings together with a brief explanation of each warning (see also section 8.1). In order to fix wrong settings you may have to close the TTTR mode dialog and return to the control panel.



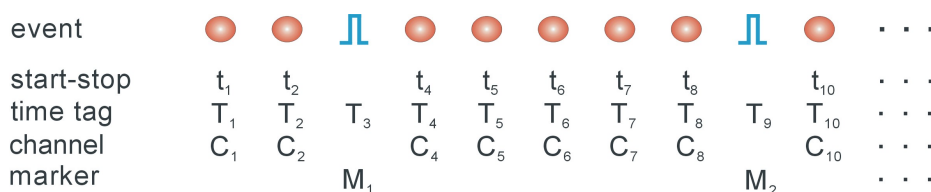
A measurement can be stopped at any time by clicking the Stop button. The data recorded up to this point will be stored in the file. When the measurement has completed, the Stop button will be grayed out (disabled). Use the Cancel button to return to the normal interactive mode. Again, this will take some time for hardware reconfiguration.

As outlined above, TTTR data collection at high rates is a demanding real-time streaming process. The hardware and software must ensure not to lose any data. In order to implement this efficiently the MultiHarp software employs multiple threads (concurrent CPU processes). A first thread continuously reads the MultiHarp's hardware FIFO and puts the retrieved data in a software queue. A second thread concurrently reads this software queue and writes the data to disk. If real-time correlation is being performed then this is done in further separate threads. User interface and interaction are handled in yet another thread. Multi-core CPUs are particularly useful here as they can run the threads in parallel rather than switching between them. There are two typical error scenarios that you may encounter in this process. The first is a situation where the first thread does not empty the hardware FIFO quickly enough and the FIFO runs full. The software then reports the error FIFO_OVERFLOW. In order to avoid this you may have to reduce the input data rate. Another error situation may result when the second thread cannot write to disk quickly enough and the software queue runs full. The software then reports the error STORAGE_QUEUE_OVERRUN. In order to avoid this you may want to check the write speed of your hard disk and see how it can be improved.

5.3.5. External Markers

Often it is desirable to synchronize TCSPC measurements with other information or processes of complex measurement tasks. In order to perform e.g., Fluorescence Lifetime Imaging, the spatial origin of the photons must be recorded as well as their timing. For this purpose one needs a mechanism to assign external synchronization information to the TCSPC data. In the special case of Fluorescence Lifetime Imaging, conventional systems use on-board memory and switch to new blocks of memory upon arrival of e.g., a pixel clock pulse. Accommodating the large amount of data generated by the 3-dimensional matrix of pixel co-ordinates and lifetime histogram bins is a serious challenge. Even with modern memory chips, this approach is limited in image size and / or number. In addition, it is expensive, and implies loss of information about the individual photon arrival times. To solve the problem in a much more elegant manner, the TTTR data stream generated by the MultiHarp can contain markers for synchronization information derived from an imaging device, e.g., a scan controller. For this purpose the control port of the MultiHarp 150 provides four TTL inputs for synchronization signals M1..M4 (see section 8.3.2).

The figure below illustrates how the external marker signals are recorded in the data stream.



Bullets denote a photon, blue pulses denote a marker signal. The external markers are treated almost as if they were regular photon event records. A special channel code allows to distinguish true photon records from marker records. Software reading the TTTR file can thereby filter out the markers e.g., for line and frame clock in imaging applications. This makes possible to reconstruct the 2D image from the stream of TTTR records, since the relevant XY position of a scanner can be determined during the data analysis. The data generated this way is nearly free of redundancy and can therefore be transferred in real-time. The image size is unlimited both in XY and in count depth. Since there are four such synchronisation signals, all imaging applications can be implemented and even other experiment control signals can be recorded. This marker scheme is a very special feature of PicoQuant's TCSPC electronics. It may be worth noting that inventing this technology enabled PicoQuant GmbH to develop the cutting edge MicroTime 200 Fluorescence Lifetime Microscope.

The TTL compatible inputs accept the synchronization signals that will be recorded as markers. The active edges of these signals can be chosen in the general settings dialog (available through the Toolbar). Both high and low state must be at least 50 ns long. The period may therefore (in principle) be as short as 100 ns but data bus throughput constraints will apply. Each marker creates an additional TTTR record, so that one must ensure not to swamp the data stream with too many marker records. In case of data bus congestion markers take precedence over photon records, so that excessive marker traffic can suppress photon records. In fast imaging applications it is therefore recommended not to use a pixel clock but a line clock only. Since each photon has a time tag, it is usually not necessary to use an additional pixel clock. Instead, virtual pixels can be defined by

subdividing the scan lines in time. The accuracy of marker timing is on the order of 50 ns. In T3 mode it can never be higher than one sync period.

A programmable marker hold-off time can be used to suppress glitches on the marker signals that some poorly designed scan hardware or cable reflections might create. The idea is that when a marker signal was detected the next (spurious) marker will be suppressed if it occurs within the hold-off time after the first detection. The hold-off time can be chosen in the software settings dialog available through the Toolbar.

5.3.6. Using TTTR Mode Data Files

For diagnostic purposes you may reload a T3 mode file into the MultiHarp software. The limitation is that you will only be able to form a histogram over the start-stop times in your T3 mode data. The time-tag information will not be used here. The MultiHarp software will recognize that you are loading a T3 mode file and how many records are contained in it. It will then prompt you for a range to use for histogramming. The histogram will go to blocks 0..N-1 of the histogram memory where N is the number of channels that were used in the T3 mode measurement. A TTTR mode file also contains all control panel settings that were active at the time of the measurement run. After loading a TTTR mode file, you will find the document title reading "Histogram from...". If you choose to save such data you will have to give it a new file name (*.phu). This is because now a histogram has been formed, and saving it with the same file name would destroy the original TTTR mode file. However, you may save the previously formed histogram as if it were obtained in normal interactive mode, to a standard MultiHarp histogram data file (*.phu).

Reloading T3 mode files serves as a quick diagnostic tool only. For T2 mode files such a feature is currently not available. Further processing or analysis of TTTR mode data must therefore be performed through external data analysis software. Such software is available from PicoQuant for a wide range of analysis tasks (under the product names of SymPhoTime and QuCoa). Further specialized analysis can be performed by dedicated custom software. If you wish to save the cost for the commercial TTTR analysis software or if you require special analysis algorithms you may want to program your own analysis software. For development of your own custom programs, please refer to the demo code for loading .phu and .ptu files. Demo source code is included on your MultiHarp installation media and will be installed by the software setup into the subfolder `filedemo`. Also see section 8.2 for the file format specifications. The paragraph below gives only an outline.

The first part of a TTTR mode file is a header with the basic setup information, similar to that of the other modes. What follows after the header is a sequence of 32 bit TTTR records. The TTTR records in the file consist of different pieces of information in groups of bits. These pieces of information must be extracted by a bit masking / shifting operation. Their specific layout is different for T2 and T3 mode. In both cases, in addition to extracting the bit fields, the software must step through the whole file and interpret the overflow records and correct the overall time axis accordingly. Details on how this can be done should be looked up in the demo programs that are installed as part of the software distribution.

5.3.7. TTTR Mode Measurements with Real-Time Correlation

An important application of the MultiHarp and its TTTR mode is Fluorescence Correlation Spectroscopy (FCS). While TTTR data collection and off-line software correlation has been used for quite some time, PCs have only recently become fast enough to calculate the correlation function "on the fly" while the data is being collected. This capability can be very useful in setting up and monitoring FCS experiments. The MultiHarp software provides such a real-time software correlator for T2 and T3 mode.

Although it collects the same type of data as regular TTTR mode, the real-time correlator has its own button on the Toolbar.



The next figure shows the TTTR mode correlator dialog.

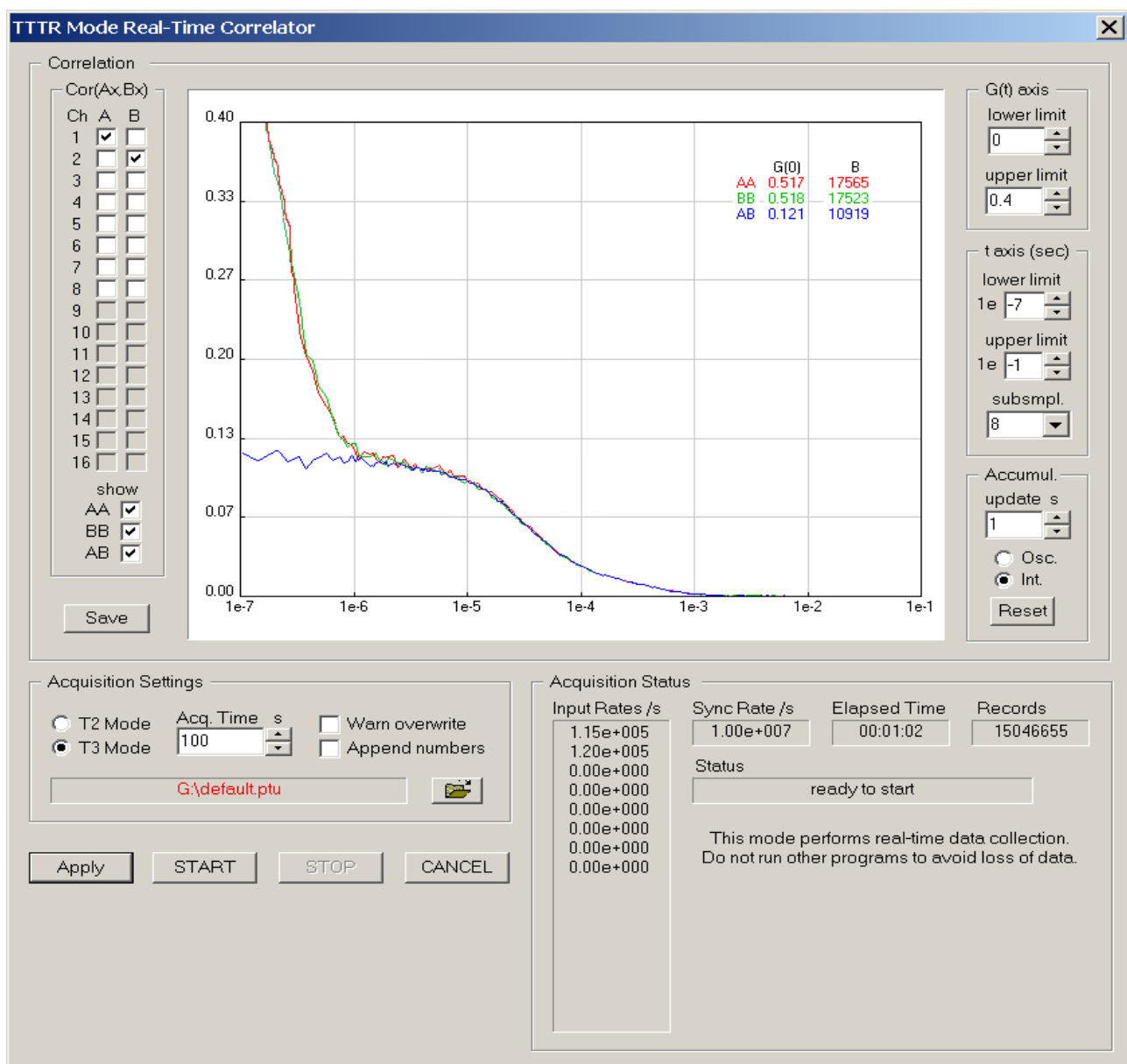
The lower section of the dialog provides the same file handling and progress indicators elements as in regular TTTR mode. Please refer to section 5.3 for details. The upper section of the dialog contains the correlator display and various control elements. The dialog section *Cor(Ax,Bx)* allows to select individual input channels for the correlation. A and B are the virtual input channels to be correlated. All input channels that are ticked in columns A and B, respectively, will be aggregated and used in the corresponding correlation channel. If your selection is empty, the heading of the section *Cor(Ax,Bx)* will turn red to indicate the problem. If you proceed anyway you will get error messages upon start.

The correlator always calculates the autocorrelations of the two virtual channels (AA and BB) as well as the cross-correlation (AB). There are three tick boxes that select which of these are shown.

The dialog section *Accumul.* allows selecting an update time for the correlation display. Furthermore it allows to select between repetitive updates (*Osc.*) and cumulative collection (*Int.*). If the latter is chosen one can manually reset the correlator by clicking the *Reset* button. Note that all these settings affect only the real-time correlator. The raw TTTR mode data will be collected continuously and completely independent from the correlator settings.

The *Save* button under the *Accumul.* section allows to save the correlator results as they were last shown. The saved result is an ASCII file with some header information. The format is self-explanatory.

Note that the real-time correlator is primarily a tool for preview during measurement setup and optimization. It does not allow to correlate data from TTTR files. If you need the correlation over the entire stream of collected data you need to make sure that the measurement runs in Integrating mode and that you do not press reset during the acquisition. If you need the correlation results you should save them here. If you need to perform further or more thorough FCS analysis on the collected data you can use the SymPhoTime 64 software from PicoQuant.



On the right hand side there are controls for the axis ranges of the correlator display. Note that they affect only the display. Collected data is always complete, independent from the axis settings. In contrast, the selection box *subsmpl.* has an effect on the correlator results. It determines how many tau points are calculated. The correlator works in a logarithmic multiple tau scheme and *subsmpl.* specifies the number of linear subsamples in each log stage. A higher number of subsamples increases the resolution of the correlation curve. Calculating more points is more time consuming and therefore may lead to lower count rate limits that can be handled. In this case you may get FIFO overruns. The default of *subsmpl.=8* is a reasonable trade-off between speed and resolution. On faster computers the setting of *subsmpl.=16* is a good choice.

Note that the starting point and spacing of the tau sampling points also depends on the time tag resolution. In T3 mode this corresponds to the sync period. In T2 mode the native resolution of the board is binned down to a time tag resolution of 25 ns in order to make the data manageable for the real-time correlation algorithm.

The correlation curve display can be shown with a grid (see general settings dialog, accessible from the toolbar of the main window). It also shows two useful figures obtained from the collected data (top right of curve window). The first figure is an approximation of $G(0)$. In classic FCS experiments this corresponds to the inverse of the number of particles in the focal volume. It is continuously updated together with the curve display, which can be useful for system adjustments, notably when using the repetitive accumulation mode (Osc.). Note that the approximation is a simple averaging over the first ten tau points. The figure B is an indicator for molecular brightness. It is also updated continuously. Note that it is also only an approximation obtained by multiplying the $G(0)$ approximation with the average count rate on both virtual correlation channels A and B. Depending on the chosen channels in A and B this may lead to figures that do not truly reflect molecular brightnesses. However, they should be useful as an adjustment aid in any case.

All other aspects of TTTR data collection with correlator preview are the same as in plain TTTR mode as described in the previous subsections. Topics such as using external markers or how to use the data files should be looked up there.

5.4. Time-Resolved Excitation and Emission Spectra

In time-resolved fluorescence research it is often of interest to observe time-dependent spectral shifts and decay changes in the context of solvent relaxation dynamics and general spectral evolution. This requires recording Time-Resolved Excitation/Emission Spectra (TRES), ideally with automated wavelength scan and TCSPC data collection under full software control.

In order to measure spectra in combination with fluorescence lifetime, the MultiHarp software provides an automated TRES measurement mode. This mode allows to control a monochromator via a stepper motor and automated collection of spectrally resolved lifetime histograms. Data is collected as in standard "Integration Mode" and saved in different blocks of memory for each wavelength. Note, however, that TRES data is always collected through input channel 1 only.

If a monochromator with appropriate stepper motor and associated drivers is installed, a specialized dynamic link library (Mono.dll) can be installed together with the MultiHarp software. The presence of this DLL will be detected by the software. In this case the button for Monochromator and TRES control on the toolbar will be enabled.



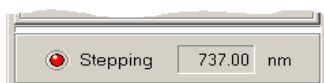
Note that most of the monochromators can be controlled only through dedicated stepper motor hardware from PicoQuant or selected vendors, usually installed as part of a complete PicoQuant spectrometer. Custom configurations can only be supported upon special request.

Clicking the monochromator button will launch the dialog for manual monochromator control and TRES setup. There you can set up parameters such as the start, step, and end of the wavelength scan. There will be error messages if the monochromator / stepper and associated drivers are not configured properly.

The monochromator used for TRES measurements is controlled through a special dialog. It shows three tabs at the top and a status panel at the bottom of the dialog. The three tabs give access to the *Manual*, *TRES*, and *Initialization* pages of the dialog.



The status panel shows the current monochromator position in *nm* and whether the monochromator is currently moving (the red "LED" labelled *Stepping* is flashing, when it moves).



The dialog is "non modal", i.e. it does not need to be closed before other windows can be accessed. It has its own icon in the Windows Taskbar, from where it can be brought back to the top, should it have been covered by other windows.

Monochromator Models

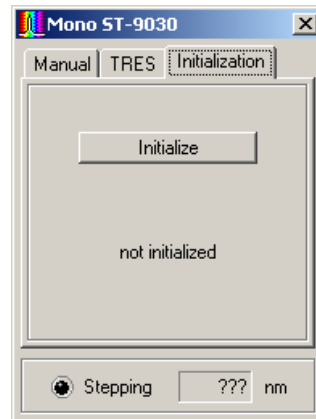
The type of the current monochromator is displayed in the title bar of the dialog. Apart from a simulation mode, the following monochromator models are supported: Sciencetech ST-9030, ST-9030DS and ST-9055, Acton Research SP-2150, SP-2155, SP-150 and SP-275. Some other Acton Research models are partially supported. Note that some monochromators can only be controlled through dedicated stepper motor hardware from PicoQuant or the monochromator supplier. Usually these are installed as part of a complete PicoQuant spectrometer. Please contact PicoQuant for details. Note also that there are more recent and superior spectrometer solutions from PicoQuant. Monochromator support for the MultiHarp software is provided for backwards compatibility only and most likely going to be phased out.

If your system is not configured for use with a monochromator, the Monochromator / TRES dialog will display the caption *Simulation*. This is for demonstration purposes only. It will work the same way as the "real" ST-9030 monochromator described below, except that after pressing the *Initialize* button, you will be prompted to decide whether the initialization should be successful or not. To continue with the demonstration, press *Yes*. Of course the simulated monochromator movements will not have any effect on the measured TCSPC histograms.

Monochromator Initialization

Before use, the monochromator must be initialized. This means, it has to seek its mechanical reference position (determined by a micro switch). Physical calibration values are kept in a monochromator configuration file (monochromator.cfg) which is generated by the system supplier at the time of delivery. Do not edit this file unless you have exact instructions for doing so.

Position initialization is required each time the software is started. It can be controlled on the *Initialization* page of the dialog. At first startup the dialog shows the state "not initialized". This means the software has no information about the current monochromator / stepper position.



The position initialization is started by clicking on the *Initialize* button. If the initialization is running, the caption of the button will change to *Stop* and the LED will start flashing. You should also hear the stepper motor. If the initialization is successful, the string "not initialized" will change to "Initialization OK", otherwise it will change to "Initialization failed!". This may happen if either the *Stop* button was pressed or an error occurred during the initialization process. In the latter case, check cabling and power supply of the system. The *Manual* and *TRES* pages will not be available unless the initialization was successfully performed. If for any reason the initialization becomes invalid (e.g., if the monochromator was turned manually and subsequently accidentally reaches the end point switch) the *Initialization* page will be activated and access to the *Manual* and *TRES* tabs will be blocked.

Manual Monochromator Control

The *Manual* page of the dialog can be used to position the monochromator manually at a given wavelength.

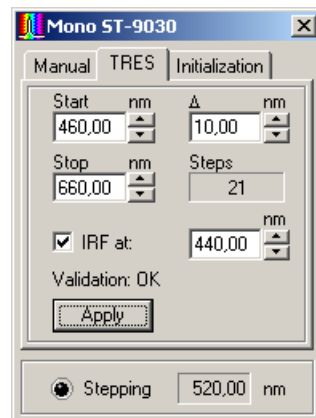


The desired position can be entered in the *Position* edit box. Clicking *Apply* or Pressing <Enter> will cause the monochromator to move to the specified position. Alternatively, the spin buttons right to the edit box can be used to modify the position. The step width of this action is defined in the Δ (delta) edit box. Pressing arrow buttons will make the change take effect immediately; entering values by keyboard require pressing *Apply* or <Enter> to show an effect.

If there is a large difference between the current monochromator position and the position entered, the movement will take some time. If you wish to interrupt this process, press the *Stop* button. The monochromator will immediately slow down and then stop. The deceleration is necessary to preserve the calibration.

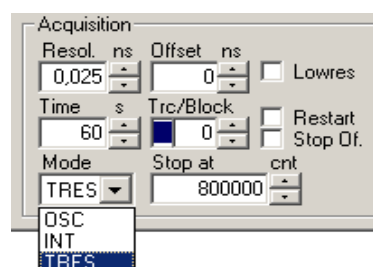
Running TRES Measurements

The *TRES* page of the dialog contains the controls used to set up an automated scanning of the monochromator. The start and stop wavelengths of the scan can be entered in the corresponding edit boxes, the Δ (delta) box defines the step width of the scan. The scan starts always exactly at the *Start* value. If the *Stop* position cannot be reached by an integer multiple of the step width, the scan stops at the last position within the start / stop range. If the start position is larger than the stop position, the scanning direction will be reversed. The maximum range and number of wavelength steps is determined by the physical characteristics of the monochromator as well as the maximum number of memory blocks to record histograms in (512). Again, pressing the spin buttons will apply changes in the corresponding edit boxes immediately; entering values manually require pressing <Enter> or the *Apply* button.



The IRF can be measured automatically as part of the scan, at a separately chosen wavelength. To do so, check the box entitled *IRF at* and enter the wavelength at which to measure the IRF in the corresponding edit box. The monochromator will go to the IRF position at the beginning of the TRES measurement. After the IRF was measured, the actual TRES scan will be performed. The IRF will always be recorded in memory block 0, while the wavelength scan always starts in block 1. Note that starting a TRES measurement will clear and subsequently overwrite all curves in memory that may have been recorded previously. If you wish to measure the IRF yourself, you must do this *AFTER* the TRES measurement. If the curves in memory were loaded from a file, the actual file on disk will not be overwritten until you save to the file (Save menu or corresponding toolbar button). Use Save As and select a new file name if you want to preserve the old file. Note that a TRES file will remain a TRES file for its lifetime. This means that after creating or re-loading such a file you can do only two things: perform a new TRES measurement (effectively overwriting the old file contents) or measure an IRF into curve 0 (overwriting only curve 0). If you want to perform any other measurement you will need to create a new file or load a non-TRES file.

The TRES measurement is selected in the MultiHarp control panel. You need to use the selection box for the measurement mode to select TRES mode (instead of OSC or INT).



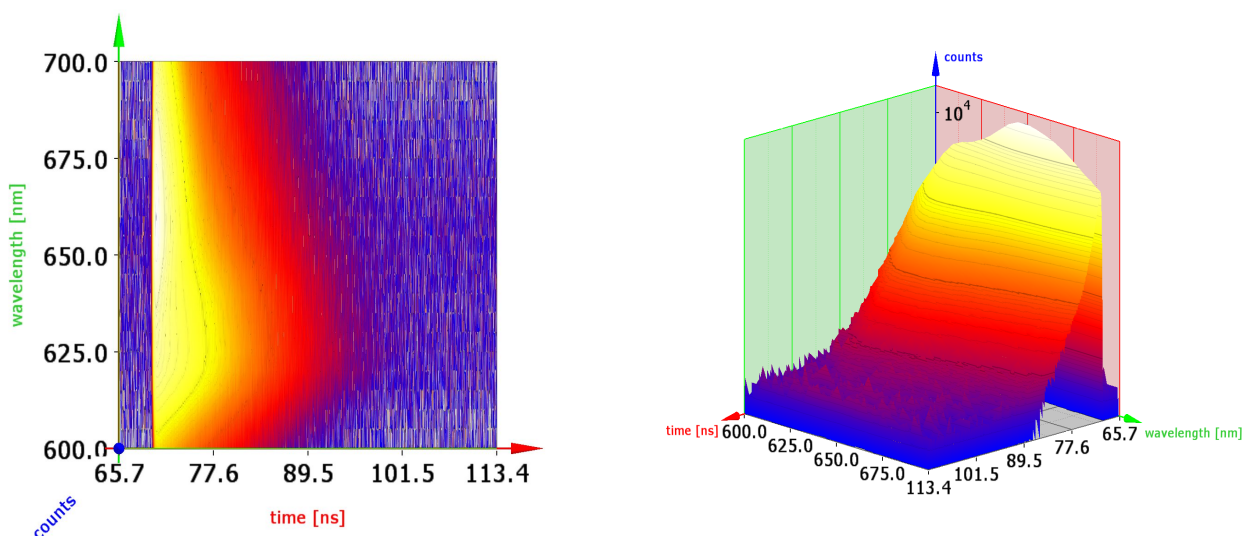
A TRES measurement can then be started or canceled as usual via the toolbar buttons for start and stop. The data acquisition will be performed like a standard “Integration Mode” measurement, while the block number is incremented for each new wavelength.



At the beginning of each TRES run, the active curve (block) will be set to 0, or to 1 if no IRF is collected. All curves that were previously collected and not saved to a file will be overwritten with new data. During a TRES run, the currently collected data will always be shown as Trace 0 (dark blue), even though the block (Trace) number is actually incremented at each wavelength step. This is to overcome the limitation of only 16 display curves being available. You can check the block number in the control panel to see the curve currently being collected. Manual entry will be disabled during the run. Also, you can watch the status bar to see the current wavelength and stepping activity. Note that for a TRES measurement the control panel setting 'Stop Of' applies as in Integration mode, while 'Restart' is meaningless in TRES mode.

TRES Data Analysis and Visualization

Having collected a complete TRES data set you can save it to a regular *.phu file. You can also inspect individual curves via the Trace Mapping dialog. You can furthermore use the software tool FluoPlot from PicoQuant to visualize and analyze the data in various 2D and 3D representations with a multitude of options for colouring, scaling, and changing view aspects in 3D. The figures below show FluoPlot visualizations from a TRES measurement of mixed oxazine dyes in ethanol.



FluoPlot is provided on your software installation media. Just run FluoPlotSetup.exe from there. If you received your MultiHarp software by download or email you may need to request FluoPlot separately. Note that the program requires a modern graphics processing unit with support for OpenGL version 1.5 or higher. Speedy handling of large data files requires sufficient system memory. Further data analysis may need to be performed by dedicated software, either custom programs or specialized solutions available from PicoQuant.

5.5. Multi-Channel Scaling

In contrast to classic TCSPC systems (based on TACs), the sync and the signal channels of the MultiHarp are completely independent. This makes it possible to allow multi-stop measurements, i.e. the detection of multiple photons between two subsequent START signals. The shorter the deadtimes of the detectors and the timing electronics are, the less these multi-stop measurements are prone to deadtime-induced photon losses. If the histogram bin width is chosen wider than the deadtime, the pile-up artifacts due to deadtime are entirely eliminated and it is possible to measure at photon rates much higher than the classic pile-up limit of TCSPC. The MultiHarp 150 can then be operated like a Multi-Channel Scaler.

The easiest way to perform multi-channel scaling with the MultiHarp is via histogramming mode. The detector is connected to one of the input channels. In addition, a reference signal at the sync input marking the start of the MCS measurement is needed (see chapter 8.3.1 for the signal specifications).




The integration time should then be set to a value significantly larger than the time bin width multiplied by the number of time bins (65536). Then start a measurement in integration (INT) mode. The resulting histogram will display the MCS curve. Please note that the time range that can be obtained with this method is limited to the time bin width multiplied by 65536. MCS measurements on longer time ranges are possible via TTTR mode. However, this will require additional software for data analysis (e.g., SymPhoTime 64).

6. Controls and Commands Reference

6.1. Main Window

The Title Bar

The title bar is located along the top of a window. To move the window, drag the title bar. Note: You can also move dialog boxes by dragging their title bars. The title bar may contain the following elements:

| | |
|---|---|
| System Menu | |
| MultiHarp – [Name of the file or dialog] | |
| <i>Minimize</i> Button |  |
| <i>Maximize</i> or <i>Restore</i> Button, resp. |  |
| <i>Close</i> Button |  |

Scroll bars

Displayed at the right and bottom edges of the MultiHarp window if a certain minimum window size is reached. The scroll boxes inside the scroll bars indicate your vertical and horizontal location in the display area. You can use the mouse to scroll to other parts of the window.

Size command

Use this command to resize the active window.

Note: The command is unavailable for already maximized or minimized windows.

Shortcut

Mouse: Drag the corners or edges of the window.
Keys: <Alt>+<Space> S

Minimize command

Use this command to reduce the MultiHarp window to an icon. Running measurements will continue.

Note: The command is unavailable for already minimized windows.

Shortcut


Mouse: Click the minimize button  on the title bar.
Keys: <Alt>+<Space> N

Maximize command

Use this command to enlarge the active window to fill the available space.

Note: The command is unavailable for already maximized windows.

Shortcut


Mouse: Click the maximize button  on the title bar;
or double-click the title bar of a non-maximized window.
Keys: <Alt>+<Space> X

Restore command

Use this command to return from a maximized or minimized window to the previous size.

Note: The command is only available for already maximized or minimized windows.

Shortcut

Mouse: Click the maximize button  on the title bar;
or double-click the title bar of the enlarged window.


Keys: <Alt>+<Space> R

Close command

Use this command to close the active window or dialog box.

Double-clicking a system menu box is the same as choosing the Close command.

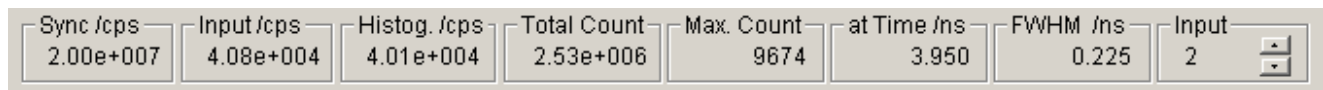
Shortcuts

Mouse: Click the close button  on the title bar.

Keys: <Alt>+<F4>
<Alt>+<Space> C

Panel Meters

At the bottom of the main window there is a set of panel meters. The meters showing units of cps (counts per second) in their title are rate meters. The leftmost rate meter shows the sync input rate. The next meter shows the channel input rate. Note the selector on the right of the panel meters. This selects the channel the rate meters (except Sync) are referring to. The other meters show the histogramming rate, the total count in the histogram, the maximum (peak) count, and the position of the maximum. They all depend on the input selector.



Note that the Input selector also has a selection option called *Sum*. In this case the meters are fed from the sum of all input channels. This is the default setting.

Also note that the rate meters have a fixed gate time of 100 ms and limited accuracy, notably at low rates. They are only a means of quick diagnostics and should not be taken for actual measurements.

6.2. Menus

6.2.1. File Menu

The *File* menu offers the following commands:

| | |
|--------------------------------------|--|
| <i>New</i> | Clears all histogram data and restores default settings. |
| <i>Open</i> | Opens an existing histogram file. |
| <i>Save</i> | Saves a histogram file. |
| <i>Save As...</i> | Saves an opened histogram file to a specified file name. |
| <i>Print</i> | Prints the currently displayed histogram. |
| <i>Page Setup</i> | Allows modifying the page layout for printing. |
| <i>Print Preview</i> | Displays the layout as it would appear printed. |
| <i>Print Setup</i> | Selects a printer and printer connection. |
| <i>1...4 <Recent Filename></i> | Opens one of the four last recently opened files |
| <i>Exit</i> | Exits MultiHarp software. |

New command

Use this command to create a blank histogram with the last default settings. You can open an existing histogram file with the *Open* command.

Shortcuts


Toolbar: 

Keys: `<Ctrl>+N`
`<Alt>+F N`

Open command

Use this command to open an existing histogram file. All control panel settings will also be restored. You can also open MultiHarp histogram files (*.thi) by double clicking them or dragging them onto the MultiHarp icon. You can revert to a blank histogram and default control panel settings with the *New* command. You can also select other file types for loading as histogram data, notably T3 mode files. Loading such files as histogram data is for diagnostics only and cannot use their full information content.

Shortcuts

Toolbar: 

Keys: `<Ctrl>+O`
`<Alt>+F O`

File Open dialog box

The following options allow you to specify which file to open:

Look in: Select (i.e. browse into) the directory in which the file that you want to open resides.

Main box In this box you see the content of the chosen directory, filtered by *Files of type*.

File name: Type or select the filename you want to open. This box lists files with the extension you select in the *Files of type* box.

Files of type Select the type of file you want to open. The default MultiHarp extension is `.thi`

Save command

Use this command to save the current histogram data to a file with current name and directory. When you save a histogram for the first time, MultiHarp displays the *Save As...* dialog box so you can name your file. This command is unavailable if a measurement is running. If you want to change the name and directory of an existing file before you save it, choose the *Save As* command.

Shortcuts

Toolbar: 

Keys: `<Ctrl>+S`
`<Alt>+F S`

Save As... command

Use this command to (re-)name the current histogram data and save. The software displays the *Save As...* dialog box so you can name your file. To save a file with its existing name and directory, use the *Save* command.

File Save As... dialog box


The following fields allow you to specify the name and location of the file you are about to save:

- Save in:* Select (i.e. browse into) the directory where you want to store the file.
- Main box* In this box you see the content of the chosen directory, filtered by *Save as type*.
- File name:* Type a new filename to save a histogram with a different name. The software automatically adds the extension you specify in the *Save as type:* box.
- Save as type:* Selects a filter on the directory chosen in the *Save in:* box. Only files that pass the filter are shown in the dialog's main box. Additionally the software adds the extension associated with the chosen filter to a given file name if it was given without extension.

Print command

Use this command to print the currently displayed histogram curves. This command presents a *Print* dialog box, where you may specify the number of copies, the destination printer, the paper orientation and other printer setup options.

Shortcuts

Toolbar: 

Keys: `<Ctrl>+P`
`<Alt>+F P`

Page Setup command

Use this command to change the print layout. This command presents the *page setup* dialog box, where you may select or deselect various items to appear in the print. You can use *print preview* to check the resulting layout.

Shortcut

Keys: `<Alt>+F U`

Print Preview command

Use this command to display the layout as it would appear when printed. The main window will be replaced with a *print preview* window in which one or two pages will be displayed in their printed format. The *print preview* toolbar offers you options to view either one or two pages at a time; move back and forth through the document; zoom in and out of pages; and initiate a *print* job.

Shortcut

Keys: `<Alt>+F V`

Print Setup command

Use this command to select a printer and a printer connection. This command presents a Print Setup dialog box, where you specify the printer and its connection.

Shortcut

Keys: <Alt>+F R

1, 2, 3, 4 command (most recently used files)

Use the numbers and filenames listed at the bottom of the *File* menu to open the last four files you closed. Choose the number that corresponds with the file you want to open.


Shortcuts

Keys: <Alt>+F 1
<Alt>+F 2
<Alt>+F 3
<Alt>+F 4

Exit command

Use this command to end your MultiHarp session. Save your data before exiting.

Shortcuts

Mouse: Click the close button  on the title bar.
Keys: <Alt>+<F4>
<Alt>+F X

6.2.2. Edit Menu

The *Edit* menu offers only one command:

| | |
|-------------|------------------------------|
| <i>Copy</i> | Copies the displayed curves. |
|-------------|------------------------------|

Copy command

Copies the currently displayed curves in ASCII format to the clipboard. This can be used to export histogram data to spreadsheet or data analysis software, e.g., the “EasyTau 2” Software. Note that only the currently selected curves within the current display limits are copied. If the curves in the display are of different resolution then all 65536 time bins will be copied. Copying data to the clipboard replaces the contents previously stored there.

The exported data is plain <Tab> separated ASCII and can be read by most commonly used spreadsheet programs or data analysis software.

This command is unavailable while a measurement is running.

Shortcuts

Keys: <Ctrl>+C
<Alt>+E C

6.2.3. View Menu

The View menu offers the following commands:

| | |
|----------------------|---------------------------------|
| <i>Toolbar</i> | Shows or hides the toolbar. |
| <i>Status Bar</i> | Shows or hides the status bar. |
| <i>Axis Panel</i> | Shows the axis settings panel. |
| <i>Control Panel</i> | Shows the control panel. |
| <i>Trace Mapping</i> | Shows the trace mapping dialog. |

Toolbar command

Use this command to display and hide the Toolbar, which includes buttons for the most common commands, such as *File Open*. A check mark appears next to the menu item when the Toolbar is displayed.

Shortcut

Keys: <Alt>+V T

Status Bar command

Use this command to display and hide the status bar, which describes the action to be executed by the selected menu item or depressed toolbar button, the current measurement activity and keyboard latch state. A check mark appears next to the menu item when the status bar is displayed. The status bar is displayed at the bottom of the MultiHarp main window.

Shortcut

Keys: <Alt>+V S

Axis Panel command

Use this command to display the axis settings panel. The result is the same as the axis panel button in the toolbar or double clicking on the histogram axes. The axis settings panel can be closed by clicking its close button.

Shortcut

Keys: <Alt>+V A

Control Panel command

Use this command to display the control panel. This command has the same effect as the control panel button in the toolbar. The control panel can be closed by clicking its close button.

Shortcut

Keys: <Alt>+V C

Trace Mapping command

Use this command to display the trace mapping dialog. This command has the same effect as the corresponding button in the toolbar. The trace mapping dialog can be closed by clicking its close button.

Shortcut

Keys: <Alt>+V M

6.2.4. Help Menu

The *Help* menu offers the following commands, which provide you assistance with this application:

| | |
|------------------------------|---|
| <i>Help Topics</i> | Offers you the contents list of topics on which you can get help. |
| <i>Help Index</i> | Offers you an index to topics on which you can get help. |
| <i>Help Search</i> | Offers you a means of full text search on all help topics. |
| <i>Activate Context Help</i> | Switches the cursor mode to “point to item” for context help. |
| <i>Check for Updates</i> | Checks for the availability of a newer software version. |
| <i>Visit Website</i> | Opens www.picoquant.com in your browser. |
| <i>Request Support</i> | Provides support details and access to the support website. |
| <i>About MultiHarp...</i> | Displays version information of the MultiHarp software. |

Note: Online help (context help) on most functions, dialogs, control items etc. is available via the F1 key.

Help Topics command

This command opens a browser offering a tree view like list of contents, and a help page. Browse through the contents with your mouse or the cursor keys. <Enter> opens / closes chapters, <Cur Up> / <Cur Down> turns the pages. The help page to the right of the browser will always be held up-to-date.

Shortcuts

Keys: <F1> (if online help was last recently opened in topics mode)
 <Alt>+H T

Help Index command

This command opens a browser offering an index list of keywords and a help page. Browse through the contents with your mouse or the cursor keys. <Enter> opens the current item, <Cur Up> / <Cur Down> navigates through the index.

Shortcuts

Keys: <F1> (if online help was last recently opened in index mode)
 <Alt>+H I

Help Search command

This command opens a browser offering full text search on words or word groups over the whole online help volume. You can enter more specific search pattern, using the operators "AND", "OR", "NEAR" and "NOT".

Shortcuts

Keys: <F1> (if online help was last recently opened in search mode)
 <Alt>+H S

Activate Context Help

Changes the mouse cursor mode to context help mode. You can obtain context sensitive help for a visual element in the GUI by simply clicking on it.

Shortcuts

Keys: <Shift>+<F1>
 <Alt>+H C

Check for Updates command

Use this command to check if newer software is available. This requires internet access. If a newer version is available it can immediately be downloaded. Note that this does not perform any installation. You still need to unzip and install the downloaded software. Do this only after reading the release notes.

Shortcut

Keys: <Alt>+H U

Visit Website command

Use this command to open www.picoquant.com in your browser. This requires web access.

Shortcut

Keys: <Alt>+H W

Request Support command

Use this command to open a form with important support details of your system and direct access to the support page on the web. The latter requires internet access. If you cannot access the web directly, please copy the support details and send them later by email to support@picoquant.com together with a precise description of the problem. If your setup is not working at all, provide at least the serial number of your MultiHarp and a precise description of the installation environment and the observed issues, including the exact wording of any occurring error messages.

Shortcut

Keys: <Alt>+H S

About MultiHarp... command

Use this command to display the copyright notice and version number of your MultiHarp software and hardware, if installed. It also provides access to the PicoQuant Web site and software updates.

Shortcut

Keys: <Alt>+H A

6.3. Toolbar

Toolbar buttons of special interest are explained in some more detail here:



Context Help

When you choose the *Context Help* button, the mouse pointer will change to an arrow with question mark. If you then click somewhere in the MultiHarp window, such as on another toolbar button, the help topic associated with it will be shown.



Axis Panel

Clicking this button opens the *Axis Panel*. This panel provides controls for the axis limits and lin / log display switching. Double-clicking the axes opens the same dialog. The dialog is non-modal, so it can remain permanently open. The panel will remember its last position when closed. When it is opened the next time it will be at that screen position again.



Data Cursor

Opens the data cursor dialog box. You can use this dialog to mark and retrieve the data values of individual histogram bins. A pair of crosshairs will be provided for marking data points with the mouse or keyboard. When the data cursor dialog is activated, the crosshairs will appear in the histogram display area. By clicking on or near a data point, the current crosshair (black) will snap to that point. At the same time a second crosshair (grey) will jump to the previously marked point. The data cursor dialog then shows the current count and time values for these points as well as the corresponding differences (deltas). The data cursor always applies to one "active" curve only. You can select this active curve at the top of the dialog where it is shown with its corresponding curve colour. While the grey crosshair normally jumps to the previous data point, you can modify this behavior by holding down the SHIFT key while clicking on curve points. The grey crosshair will then remain at its previous position. This allows finding a particular point while keeping the other one fixed.

Another advanced mode of operation of the data cursor is possible via the LEFT and RIGHT arrows of the keyboard. Pressing these keys will direct the black cursor to the next left or right point in the curve. Again, the behavior of the marker for the previous point (grey cross) can be controlled via the SHIFT key.



Control Panel

Launches the MultiHarp *Control Panel*. If the control panel is already open, subsequent clicking of this button will just make the control panel the active window. The dialog is non-modal, so it can remain permanently open. The panel will remember its last position when closed. When it is opened the next time it will be at that screen position again, even in the next MultiHarp session.



Trace Map

Launches the *Trace Mapping* dialog. The MultiHarp software can measure histograms in up to 512 memory blocks. Out of these, up to 16 curves can be displayed. The trace map dialog is used to select the curves to display. Tick the individual "Show" boxes to display a curve. Select the number of the memory block you wish to map the individual display curve to. Choose the active memory block you wish to use for the next measurement in the MultiHarp control panel.



TTTR Mode

Opens the TTTR mode dialog box. Use this dialog to enter the acquisition time and the destination file for a TTTR mode run. Make sure all other measurement parameters have been set and tested in interactive mode before entering TTTR mode.



TTTR Real-Time Correlator

Opens the TTTR mode real-time correlator dialog box. Use this dialog for FCS preview during a TTTR mode run. Make sure all other measurement parameters have been set and tested in interactive mode before entering TTTR correlator mode. Changing to this mode takes a few seconds for reconfiguration of the hardware. See also section 5.3.7.



Monochromator / TRES

In order to measure Time-Resolved Excitation/Emission Spectra (TRES), the MultiHarp software provides a TRES measurement mode, that allows to control a monochromator via stepper motors and automated collection of spectrally resolved lifetime histograms. Clicking this button will launch the dialog for manual monochromator control and TRES setup. There you can set parameters such as the start, stepping, and end of the wavelength scan. There will be appropriate error messages if the monochromator / stepper and associated drivers are not configured properly.



General Settings

Opens the software settings dialog box. Use this dialog to change standard settings of the MultiHarp software. Notably these are: *Display rate* (0.1 to 1s), *Draw mode* (lines, stairs), *Grid* checkbox, *Prompt overwrite* (warning before overwriting existing data), selective disabling of *Warnings* and *TTTR Marker Settings*. The control connector of the MultiHarp 150 provides TTL inputs for synchronization signals. The markers can be enabled or disabled as well as recorded at either the rising or falling edge of the corresponding TTL signal. The active edges can be chosen here. There is also a programmable hold-off time to suppress glitches on the marker signals. The dialog also allows selective disabling of warnings that you do not wish to receive. All settings will be kept in the Windows registry and will be retrieved at the next program start. They are stored individually for each Windows user profile.



White Rabbit Settings

Opens the White Rabbit dialog. Use this dialog to configure and establish a White Rabbit link for clock synchronization. White Rabbit allows time transfer and synchronization over fiber based Ethernet with sub-ns accuracy (for more information see https://en.wikipedia.org/wiki/The_White_Rabbit_Project). Note, however, that White Rabbit support is still basic, to some extent simply because White Rabbit as such is still evolving. Currently the MultiHarp 150 supports only basic White Rabbit functionality, notably clock synchronization and time transfer between two MultiHarp devices where one device is White Rabbit master and the other is White Rabbit slave. Similarly some other White Rabbit devices running as a master can act as the clock source of the MultiHarp or the MultiHarp can act as a master and clock source for some other White Rabbit device. Not all of this has yet been tested. White Rabbit support is expected to be improved with future software and firmware updates. Please contact us with your requirements if you are interested in improvements or extensions of this product feature.

6.4. Control Panel

The control panel consists of several sections containing edit boxes and other controls for related parameters. These are described in the following subsections. Note that the settings of the control panel as well as the position of the control panel on the screen will be stored in the registry and retrieved at the next program start. When you load a MultiHarp data file the settings of the control panel will change according to the settings in that file. This can be used as a means of reverting to a standard setting for the given experiment.

6.4.1. Sync–Input / Trigger Out

The settings in this group configure the behaviour of the Sync input and the Trigger output. See section 5.1 “Setting Up the Input Channels” to get started.

Sync Trigger Edge edit box and spin control

Here the trigger edge (rising/falling) for the Sync input can be set. The value 0 stands for falling, 1 stands for rising edge. Type the value as an integer in the edit box and press `<Enter>` or click on *Apply*. Alternatively, use the spin control (next to it) to increment / decrement the current value. In this case changes take effect immediately without pressing `<Enter>`.

Sync Trigger Level edit box and spin control

Here the trigger level for the Sync input can be set. Units are millivolts (mV), the permitted range is -1200 to 1200. Type the value as an integer in the edit box and press `<Enter>` or click on *Apply*. Alternatively, use the spin control (next to it) to increment / decrement the current value. In this case changes take effect immediately without pressing `<Enter>`.

Sync Offset edit box and spin control

Shifts the relative timing of Sync and photon events and is designed to compensate optical and electrical delays (due to differences in cable lengths and optical paths), e.g., in order to shift your fluorescence decay within the sync frame so that it is not truncated. This feature completely eliminates the need for adjustable delay boxes.

Units are picoseconds (ps), the permitted range is from -99,999 to +99,999 ps. Type the value as an integer in the edit box and press `<Enter>` or click on *Apply*. The offset value can be entered in steps of picoseconds but note that the actual step is one time bin according to the instrument's resolution (80 ps). Alternatively, use the spin control next to the edit box to increment/decrement the current value. In this case changes take effect immediately without pressing `<Enter>`.

Sync Tdead edit box and spin control

Tdead is a programmable dead-time for suppression of afterpulsing artefacts of some detectors. On the sync input it is rarely needed. Leave it at 0 if you work with a sync signal from a laser.

Units are nanoseconds (ns), the permitted range is from 0 to 160 ns. Note that Tdead = 0 does not really mean zero deadtime, instead, the input will then operate at its shortest (native) dead-time of about 650 ps. When a dead-time >0 is set then the timing hardware will suppress events falling into the dead-time. This is only a suppression of further processing and does not prevent the TDC to go into another (native) dead-time.

Type the value as an integer in the edit box and press `<Enter>` or click on *Apply*. The value can be entered in steps of nanoseconds but note that the actual step is approximated only to the instrument's resolution (80 ps). Alternatively, use the spin control next to the edit box to increment/decrement the current value. In this case changes take effect immediately without pressing `<Enter>`.

The programmable dead-time is only available with suitable firmware (“gateway”), which must be version 0.8 or higher. Devices shipped after September 2019 will have it by default. Otherwise the edit box for Tdead will remain grayed out. Older devices can be upgraded. An updater tool for version 0.8 is provided as part of the distribution media in the folder `gateway_update`. See `readme.txt` in that folder for instructions.

Sync Divider edit box and spin control

Here the programmable divider of the Sync input can be set. This allows to reduce the sync input rate so that the period is at least as long as the dead time. This is required for fast sync sources (≥ 78 MHz). Internal logic determines the sync period and re-calculates the sync signals that were divided out. It should be noted that this only works with stable sync sources that provide a constant pulse-to-pulse period. All fast laser sources known today meet this requirement within an error of a few picoseconds. Set the divider only as large as necessary. With random and low rate signals (< 1 MHz), the divider must be set to 'None'.

Trigger Output Period edit box and spin control

The trigger output can be used to trigger a light source. Here the period of this output can be set. The unit is microseconds. The allowed range is 0.1 μ s to 1.6 s.

Trigger Output Force On check box

Here the trigger output can be switched on regardless of whether a measurement is running.

Trigger Output Auto On check box

Here the trigger output can be set to turn on only when a measurement is running. This can reduce bleaching.

6.4.2. Inputs 1..4, 5..8, 9..12, and 13..16

The settings in these dialog tabs configure the behavior of the input channels. See section 5.1 "Setting Up the Input Channels" to get started. The MultiHarp 150 can have 4, 8 or 16 input channels. Dependent on how many channels the device has, one or more tabs with control elements for the channels will be shown. The specific controls for each channel are described below.

Trigger Edge edit box and spin control

Here the trigger edge for the input channel can be set. The value 0 stands for falling, 1 stands for rising edge. Type the value as an integer in the edit box and press `<Enter>` or click on *Apply*. Alternatively, use the spin control (next to it) to increment / decrement the current value. In this case changes take effect immediately without pressing `<Enter>`.

Trigger Level edit box and spin control

Here the trigger level for the respective input channel can be set. Units are millivolts (mV), the permitted range is -1200 to 1200. Type the value as an integer in the edit box and press `<Enter>` or click on *Apply*. Alternatively, use the spin control (next to it) to increment / decrement the current value. In this case changes take effect immediately without pressing `<Enter>`.

Offset edit box and spin control

Shifts the relative timing of photon events and is designed to compensate optical and electrical delays (due to differences in cable lengths and optical paths), e.g., in order to shift your fluorescence decay within the sync frame so that it is not truncated. This feature completely eliminates the need for adjustable delay boxes.

Units are picoseconds (ps), the permitted range is from -99,999 to +99,999 ps. Type the value as an integer in the edit box and press `<Enter>` or click on *Apply*. The offset value can be entered in steps of picoseconds. Alternatively, use the spin control next to the edit box to increment/decrement the current value. In this case changes take effect immediately without pressing `<Enter>`.

Tdead edit box and spin control

Tdead is a programmable dead-time for suppression of afterpulsing artefacts of some SPAD detectors. Otherwise it is rarely needed. Leave it at 0 unless you have good reasons.

Units are nanoseconds (ns), the permitted range is from 0 to 160 ns. Note that $T_{dead} = 0$ does not really mean zero deadtime, instead, the input will then operate at its shortest (native) dead-time of about 650 ps. When a dead-time >0 is set then the timing hardware will suppress events falling into the dead-time. This is only a suppression of further processing and does not prevent the TDC to go into another (native) dead-time.

Type the value as an integer in the edit box and press `<Enter>` or click on *Apply*. The value can be entered in steps of nanoseconds but note that the actual step is approximated only to the instrument's resolution (80 ps). Alternatively, use the spin control next to the edit box to increment/decrement the current value. In this case changes take effect immediately without pressing `<Enter>`.

The programmable dead-time is only available with suitable firmware ("gateway"), which must be version 0.8 or higher. Devices shipped after September 2019 will have it by default. Otherwise the edit box for T_{dead} will remain grayed out. Older devices can be upgraded. An updater tool for version 0.8 is provided as part of the distribution media in the folder `gateway_update`. See `readme.txt` in that folder for instructions.

6.4.3. Acquisition

Resolution edit box and spin control

Use this set of input controls to specify the time resolution. Units are picoseconds (ps). Possible choices are the device's base resolution (80 ps) and successive multiples of two. Type the desired resolution value as an integer in the edit box and press `<Enter>` or click on *Apply*. Alternatively, use the spin control (next to it) to increment / decrement the current value. In this case, changes take effect immediately without pressing `<Enter>`. In case of entering values other than valid resolutions, the next suitable resolution step is chosen automatically.

There are always $2^{16} = 65536$ time bins in one histogram. With the chosen resolution, the respective time range covered is $65536 \cdot \text{Resolution}$. The choice of range must be a compromise between resolution and time span covered. The smallest range offers the best resolution and the shortest span (vice versa for the largest range). For high sync rates the highest resolution is usually most appropriate, since the smallest range still covers the full sync period. For lower sync rates the histogram range may be too small to cover the full sync period. The decay curve region of interest may therefore lie outside the acquisition window of 65536 time bins. Apart from switching to a lower resolution it is possible to shift the acquisition window relative to the sync frame by means of the offset. Note that working with very long time spans (low sync rates) at high count rates also requires long acquisition times to minimize noticeable "steps" in the acquired histograms.

Acquisition Time edit box and spin control

Set the desired measurement time here. Units are seconds (s). The permitted range is 0.001 to 360,000 s in steps of 0.001 s. Type the desired value in the edit box and press `<Enter>` or click on *Apply*. Alternatively, use the spin control (next to it) to increment / decrement the current value. In this case, changes take effect immediately without pressing `<Enter>`. The default increment / decrement per mouse click is logarithmic.

Offset edit box and spin control

For high sync rates the highest resolution is usually most appropriate, since the smallest range still covers the full sync period. The offset should in these cases always be set to 0.

For lower sync rates the histogram range may be too small to cover the full sync period. The measurement region of interest may therefore lie outside the acquisition window of 65536 histogram bins. Apart from switching to a lower resolution it is possible to shift the acquisition window relative to the sync frame by means of setting the offset > 0 .

Use this set of input controls to specify the desired offset. Units are nanoseconds (ns). Possible offsets are in the range from 0 to 100,000,000 ns. Type the value as an integer in the edit box and press `<Enter>` or click on *Apply*. The offset value can be entered in steps of nanoseconds but will internally be rounded to the nearest multiple of the base resolution (80 ps). Alternatively, use the spin control next to the edit box to increment/decrement the current value. In this case changes take effect immediately without pressing `<Enter>`.

Internally the offset is subtracted from each start-stop measurement before it is used to address the histogram bin to be incremented. Therefore, increasing the offset means shifting the signal on the screen to the left, towards earlier times.

Note that the offset has no effect on the relative timing of laser pulses and photon events. It merely shifts the region of interest where data is to be collected. The relative timing of laser pulses and photon events can only be controlled by means of cable delays and the input offsets described in sections 6.4.1 and 6.4.2. The latter may be necessary in order to shift your fluorescence decay within the sync frame so that it is not truncated.

Trc./Block trace colour indicator, edit box and spin control

The MultiHarp software can measure and store histograms in up to 512 memory blocks. Out of these, up to 16 curves can be displayed and one "active block" can be used for measurements. Choose the active memory block you wish to use for the next measurement here. The trace mapping dialog is used to select the curves for display. Make sure the curve you are using is switched on (*shown*). You can open this dialog directly by clicking on the trace color indicator. The color indicator shows the trace colour the chosen block is currently mapped to. If it is a solid square, the curve is mapped and shown. If it is mapped but not shown, the indicator shows a small striped square. If the curve is not even mapped for display the indicator remains white. Make sure not to overwrite existing data in a block that was used before. In order to warn you that a trace is used, the heading of the Trc/Block selector will turn red.

Restart check box

If this box is ticked (checked) the measurement will automatically restart after the acquisition time has elapsed. Toggle the current setting with a mouse click. The setting is without effect in TRES mode.

Stop on Overflow check box

If this box is ticked (checked) the measurement will stop on overflow of any histogram bin. The overflow limit is set by the "Stop At" parameter. Toggle the current setting with a mouse click.

Mode selection box

Here you can select between three different acquisition modes:

Oscilloscope Mode:

In oscilloscope mode the acquisition and display, once started, repeats at intervals given by the current acquisition time setting. The histogram accumulation always starts from scratch. This is useful for monitoring fast changes during optical alignment etc. Note that in this mode you may see nothing for a long time if you set the acquisition time to a high value. Typically one would not set an acquisition time of more than 1 second in this mode.

Integration Mode:

As opposed to oscilloscope mode, the histogram acquisition in integration mode is not reset to zero with each display refresh. The histogram continues to grow while the display is updated every 0.1 to 1 seconds. The update rate is determined by the refresh rate value selected in the *General Settings Dialog*. Acquisition can be manually started and stopped, additionally the option "Stop on Overflow" will stop the acquisition when the maximum count of any histogram bin overflows. The option "Restart" will cause the acquisition to start again after the acquisition time has elapsed. This is similar to oscilloscope mode but allows watching the histogram grow if the acquisition time is fairly long.

TRES Mode:

Selects the measurement mode TRES (Time-Resolved Excitation/Emission Spectra). This requires a monochromator and corresponding driver hardware / software to be installed. See section 5.4.

Stop At edit box and spin control

Here the stop count level for histogramming can be set. The setting is only meaningful if the check box "Stop Off" is active. Type the value as an integer in the edit box and press <Enter> or click on *Apply*. Alternatively, use the spin control (next to it) to increment / decrement the current value. In this case changes take effect immediately without pressing <Enter>. The minimum is 0. If the entered value exceeds the allowed maximum of 4,294,967,295, an error message appears. When using the spin control, exceeding the maximum is not possible. Instead of incrementing the value further, the system beeps as a warning.

6.5. Axis Panel

The controls of the *Axis Panel* are used to customize the main window's curve display. All mouse actions on the spin controls on this dialog result in instant modification of the display. If you choose to alter the values in the edit fields by keyboard, you have to finalize your changes by pressing `<Enter>` or clicking the *Apply* button.

The controls are grouped as follows:

6.5.1. Time Axis Group

Note that the time axis labelling is precise in terms of relative information only. At offset=0 the absolute time difference between the inputs is close to the shown times but may be off by some tens of picoseconds. Offsets >0 change the placement of the acquisition window and the true time differences are then off accordingly.

Minimum edit box and spin control

Here the starting value of the displayed time axis can be set. Units are nanoseconds (ns). The minimum is 0. If the entered value exceeds the current time axis maximum, an error message appears. When using the spin control exceeding the current time axis maximum is not possible. Instead of incrementing the minimum value further, the system beeps as a warning. Keep in mind the notes on time axis interpretation given in the paragraph *Time Axis Group* above. Also note that the increment upon using the spin buttons grows with the current value. This permits fast changes across the entire range.

Maximum edit box and spin control

Here the end value of the displayed time axis can be set. Units are nanoseconds (ns). If the entered value becomes smaller than the current time axis minimum, an error message appears. When using the spin control, violating the current time axis minimum is automatically prevented. Instead of decrementing the minimum value further, the system beeps a warning. Keep in mind the notes on time axis interpretation given in the paragraph *Time Axis Group* above. Also note that the increment upon using the spin buttons grows with the current value. This permits fast changes across the entire range.

The time axis settings will determine the number of time bins exported via clipboard copy / paste. Only the bins within the displayed time range will be used.

6.5.2. Count Axis Group

Minimum edit box and spin control

Here the starting value of the displayed count axis can be set. Units are counts. If the minimum is set to 0 in logarithmic mode, the resulting minimum as displayed is 10^{-1} . If the entered value exceeds the current count axis maximum, an error message appears. When using the spin control, exceeding the current count axis maximum is automatically prevented. Instead of incrementing the minimum value further, the system beeps a warning. Note that the increment upon using the spin buttons grows with the current value. This permits fast changes across the entire range.

Maximum edit box and spin control

Here the upper end of the displayed count axis can be set. Units are counts. If the entered value becomes smaller than the current time axis minimum, an error message appears. When using the spin control, violating the current count axis minimum is prevented automatically. Instead of decrementing the minimum value further, the system beeps a warning. Note that the increment upon using the spin buttons grows with the current value. This permits fast changes across the entire range.

Lin / Log radio buttons

These radio buttons change the scaling of the count axis from linear to logarithmic and vice versa. Despite the fact that in logarithmic scale the display will always show whole powers of ten, the change of the scaling is applied without modifying the minimum / maximum range.

6.6. Trace Mapping Dialog

The MultiHarp software can measure and store histograms in up to 512 memory blocks. Out of these, up to 16 curves can be displayed at the same time. The trace mapping dialog is used to select the curves to display. Tick the individual boxes 'Show' to display a curve. Select the number of the memory block you wish to map the individual display curve to. The Trace Mapping Dialog also provides some statistics on each curve. These items are:

| | |
|-------------------|---|
| <i>FWHM</i> | The Full Width Half Maximum of the curve peak (usually for IRF traces) |
| <i>Max Count</i> | The count in the highest point of the curve |
| <i>At Time</i> | The time corresponding to the histogram bin where <i>Max Count</i> occurred |
| <i>Resolution</i> | The time bin width of the curve |

Furthermore there are several buttons:

| | |
|--------------------------|---|
| <i>Details</i> | Can be clicked to see more curve information |
| <i>All</i> | Can be clicked to tick all traces as shown |
| <i>0..15</i> | Can be clicked to set the default mapping of trace 0..15 to block 0..15 |
| <i>Clear (trash can)</i> | Can be clicked to delete the contents of individual blocks |

The trace mapping dialog can be launched from the corresponding button on the toolbar as well as through the trace colour indicator on the control panel.

6.7. General Settings Dialog

Use this dialog to change standard settings of the MultiHarp software. Notably these are: *Prompt overwrite* (warning before overwriting existing data) and *TTTR Marker Settings*. The control connector of the MultiHarp 150 provides TTL inputs for synchronization signals. The markers can be enabled or disabled and recorded at either the rising or falling edge of the corresponding TTL signal. The active edges can be chosen here. All settings will be kept in the Windows registry and will be retrieved at the next program start. They are stored on a per user basis.

The dialog's controls are grouped as follows:

Display Group

| | |
|------------------------|--|
| <i>Display Rate /s</i> | Sets the time interval between display updates (from 0.1 to 1s). |
| <i>Draw mode</i> | Switches between different curve draw-modes (Lines / Stairs). |
| <i>Grid</i> | Checked: Shows a light grey grid on the curve display. Unchecked: The axes of the curve display are marked with ticks |

File Saving Group

| | |
|-------------------------|---|
| <i>Prompt overwrite</i> | Activate this check box if you wish to be prompted before saving data over existing data in a file (recommended). |
|-------------------------|---|

Warnings Group

Here you can selectively enable / disable individual warnings. Activate the check box for each warning you wish to receive. See section 8.1 for details.

TTTR Marker Settings Group

Here you can set the signal specifications for external marker signals used in TTTR mode (T2 and T3 mode). The settings for the different markers are independent from each other. The following table applies to each of the four markers:

| | |
|-------------------------|---|
| <i>enable</i> | Check this if you want the marker to be included into the TTTR data stream. |
| <i>rising / falling</i> | Radio buttons to identify the active edge of the signal. |

Clock Source Group

Here you can select the MultiHarp's clock source. Normally it runs on its own internal crystal clock. In cases where clock synchronization with other devices is required it is possible to use an external source, e.g., another MultiHarp or an atomic clock. The latter is of interest when a more accurate clock is required. In each case the clock source must deliver an industry standard 10 MHz clock signal. When remote synchronization is required the best option is White Rabbit. In this case the White Rabbit link must have been established before switching to White Rabbit as a clock source. Note that any change of the clock source requires re-initialization of the MultiHarp. Also note that any disconnection from or interruption of the external clock will cause the MultiHarp to fail and report a system error. To recover from this error state the device must be re-initialized after the clock source is stable again.

6.8. White Rabbit Dialog

This dialog allows configuration and control of the MultiHarp's built-in White Rabbit PTP Core (WRPC). This core runs autonomously in the MultiHarp's FPGA and is not normally required to use the MultiHarp's regular TC-SPC and time tagging functionality. However, once a White Rabbit connection has been established it is possible to switch the MultiHarp's clock source over to White Rabbit and then time tagging can be performed synchronous to the White Rabbit clock and remote device(s) thereby synchronized to the same timing reference.

For general information on White Rabbit please refer to the website
<https://www.ohwr.org/projects/white-rabbit>.

For information on the White Rabbit PTP Core please refer to the page
https://www.ohwr.org/projects/wr-cores/wiki/WRpc_core.

For details on the specific version 4.0 of the core currently used in the MultiHarp please refer to
<https://www.ohwr.org/projects/wr-cores/wiki/wrpc-release-v40>.

6.8.1. Description of Controls

The White Rabbit Dialog provides three groups of controls that are described in the following subsections.

White Rabbit EEPROM Data

In order to use White Rabbit in the MultiHarp it is first necessary to provide the WRPC with some configuration data. This configuration data is stored in Electrically Erasable Programmable Read-Only Memory (EEPROM) and the controls in this group serve the purpose of editing, storing, and retrieving it.

The topmost set of controls labelled **MAC** is used to assign a Media Access Control (MAC) address to the MultiHarp's WR network port. The MAC is essentially a string of 6 bytes and must be unique within your network. If you are connecting to a non-private network then it should also be globally unique*. The button `Read` will retrieve the MAC address from EEPROM and place it as a hexadecimal number in the edit field on the left. If there is no MAC address in EEPROM you may want to enter one in the edit field (as a hexadecimal number) and then click the button `Write` in order to store it in EEPROM. The status field on the right will show success or failure information regarding the last read or write activity.

The next set of controls further down, labelled **Init Script** is for configuration of an initialization script that runs when the WRPC starts (i.e. when the WR link is switched on, see below). For details of the script syntax please refer to the WRPC documentation. The procedure of reading and writing the script from/to EEPROM is the same as described for the MAC address above.

* Official and guaranteed unique MAC addresses can be obtained from IEEE (<http://www.ieee.org>)

The next set of controls further down, labelled **SFP DATA** is for configuration of the Small Form-factor Pluggable transceiver (SFP) module(s) being used. The WR socket at the MultiHarp's front panel is a receptacle for an SFP module converting between electrical and optical signals. The SFP modules require calibration and the group of controls described here allows for depositing the calibration data so that the WRPC can use it. The controls and procedures for editing, reading and writing are the same as described for the other two control groups above. The EEPROM provides space for calibration data of up to 4 SFP modules. This corresponds to 4 lines in the edit box on the left. The syntax for each line is

```
SFP-Module-name Delta-Tx Delta-Rx Alpha
```

where the SFP-Module-name is an ASCII string given by the module manufacturer and the following three calibration values are signed decimal numbers. All items are separated by spaces. Not all lines need to be populated. The specific calibration parameters must be obtained by a calibration procedure according to the WRPC manual. If you order SFP modules from PicoQuant they will be shipped as a pair with the following names and calibration values (valid Oct. 2018):

```
AXGE-1254-0531    218466    218726    63293704
AXGE-3454-0531    217080    217320    -60056437
```

You can copy these two lines directly into the edit field and write them to EEPROM. Should other models be shipped at a later time the calibration data will be provided with the modules. If you obtain SFP modules from other sources you will have to perform the calibration on your own.

WR Status

This group of controls provides selected status information of the WRPC. It may be retrieved manually by clicking the **Refresh** button. It may also be retrieved periodically by checking the tickbox **Auto**. Note that the status information is retrieved from the WRPC's console output, which is available only when the WRPC is running and has received the command `gui`.

Start/Stop/Restart

This group of controls provides a drop-down menu for mode selection and buttons for manual control of the WRPC. The usage of these buttons will be explained further down as part of the description of the related WR initialization procedures.

WR Console Output

The WRPC is internally running an embedded software that provides a command console that in principle be accessed like using a terminal. The current implementation in the MultiHarp firmware and software does not allow full access to this console. However, the group of controls described here provides read access to the console output. If the tickbox **Auto-Refresh** is checked the WR console output is periodically refreshed. Note that the WRPC's console output is providing meaningful status information only when the WRPC is running and has received the command `gui` which starts the WRPC monitor.

6.8.2. Setting up White Rabbit Connections

As outlined above, using White Rabbit as a clock source for the MultiHarp requires establishing a WR connection. The following describes the necessary steps to set up such a connection by the example of a point to point WR connection between two MultiHarp devices.

As a first step it is always necessary to switch the WR link on (starting the embedded WRPC software). This is done by clicking the button **Link On** in the WR dialog of both devices. In order to see if this is successful it is advisable to have the automatic status refresh enabled. It may take a few seconds until the link is established. Now you can select the WR mode of one device as master and the other as slave. Then click the button **Set Mode** in each device's WR dialog. Observe the status display and wait until the PTP status switches to MASTER and SLAVE respectively. The console output should now say "Locked Calibrated" in green letters on both sides and on the slave side the servo state should eventually switch to "TRACK_PHASE". The WR connection is now established and you can inspect the console output, e.g., to check the timing accuracy between the two devices or other connection statistics. For details please refer to the WRPC manual.

After the WR connection is established it is possible to set the current time of the master side as the common time for master and slave. This can be done by clicking the button *Set Time* at the master. Note that this time is obtained from the operating system (as UTC). This is only as accurate w.r.t. true UTC as the accuracy of the master's Windows clock setting. More accurate time can be obtained by using the Grand Master mode which, however, requires an accurate time source such as a GPS timing receiver, an atomic clock or similar, connected to the MultiHarp via the 10MHz clock reference and PPS inputs at the back of the housing. Note however, that such functionality is not yet conveniently supported by the MultiHarp software. Serious work in this direction should preferably use the MultiHarp programming library and custom software.

When the WR connection is established and (optionally) the WR time has been set, the WR dialog can be closed and WR can be selected as the reference clock source for the MultiHarp. Note that the choice of master versus slave in the clock source selection must match the role of master/slave of the WR connection.

For regular use of the WR connection it is rather cumbersome to perform the described connection procedure manually each time. Instead, it is possible to place an appropriate initialization script in master and slave so that it is automatically executed upon power-up. An example for the slave side may look as follows:

```
ptp stop
sfp detect
sfp match
mode slave
ptp start
gui
```

The only difference for the master side is setting `mode master` (or `mode gm` for grandmaster). Please note that the last line `gui` is not strictly necessary in a generic WRPC node. However, due to the MultiHarp's current implementation it is required because the WRPC monitor output is used to retrieve status information for the related programming library functions and the status information provided in the WR dialog. For more details on the script commands please refer to the WRPC documentation.

So far, only a point to point WR connection between two MultiHarp devices was discussed. In principle it is also possible to connect a MultiHarp with some other WR node, however, it has not yet been possible to perform the necessary tests except with one generic device (SPEC board from Seven Solutions). Similarly, the MultiHarp should also work in a true WR network with multiple nodes, however, at the time of this software release this has not yet been tested. In particular, currently only connections by MAC address have been tested and IP addresses are not assigned. Please contact us if you wish to operate the MultiHarp in a full blown network. Furthermore, some advanced uses of WR such as remote experiment control and joint action of multiple devices are not yet fully implemented. We look forward to collaborate with interested users in order to extend and improve the MultiHarp's WR functionality in future software/firmware versions. Please get in touch.

6.9. About MultiHarp... Dialog

This dialog provides version information on the MultiHarp software and hardware, the latter only if it is connected and operational. It can be opened via the Help menu or via the toolbar. The button *Request Support* opens a small text viewer window that provides a listing of MultiHarp hardware and software versions that you can copy and paste into your support enquiry. Note that this information is very important for adequate support. Support requests without this information cannot be processed and will be delayed by return questions for this information. If your system is not functional at all, the minimum information you must provide for support is the serial number of your MultiHarp. It can be found at the back of the housing. The dialog also provides buttons for links to relevant web pages and a button for checking for and downloading software updates. Note that downloading a software update does not automatically install it. Typically the downloads contain zip files that must be unpacked and installed manually. Also note that it may not always be advisable to blindly install the latest software. This is the case especially when a user or his/her laboratory have developed custom software that relies on a certain file format. Updates may break such compatibility. Therefore, always carefully read the release notes before installing.

6.10. Title and Comment Editor

You can use this dialog to edit the file title / comment. It can be opened via double-click on the title, via the *File* menu or via the *Print Preview* Toolbar. The text you enter here will be stored in the data file. The first line will be displayed as the file title above the histogram display area. The text you can enter here is limited to 4 lines and 255 characters. Upon loading of a data file, the title and comment will also be retrieved. It will also be included in prints, if the corresponding check mark is set in the page setup dialog box available through the *File* menu.

6.11. Print Preview Dialog

Use this Dialog to preview the layout as it would appear when printed. When you choose this command, the main window will be replaced with a print preview window in which one or two pages will be displayed in their printed format. The print preview dialog offers the following options:

Print

Bring up the print dialog box to start a print job.

Next Page

Preview the next printed page.

Prev Page

Preview the previous printed page.

One Page / Two Page

Preview one or two printed pages at a time.

Zoom In

Take a closer look at the printed page.

Zoom Out

Take a larger look at the printed page.

Page Setup

Change the layout of the printed page.

Close

Return from print preview to the editing window.

7. Problems, Tips & Tricks

7.1. PC Performance Issues

The MultiHarp device and its software interface are a complex real-time measurement system requiring appropriate performance both from the host PC and the operating system. This is why a fairly modern CPU and sufficient memory are required, along with a recent USB 3.0 or USB 3.1 host controller. The screen resolution should be at least 800x600. At least a 2 GHz dual core (better quad core) processor, 4 GB of memory and a fast hard disk (preferably SSD) are recommended.

In order to maintain correct interaction between the measurement hardware, the display of histogram curves and user input, the operating system's message passing mechanism is used. It is recommended not to overload the system by running other processes in the background while measuring with the MultiHarp. The PC's own occasional network activity should be no problem but running the machine e.g., as a server for other PCs is not recommended. In principle any kind of background activity is allowed. However, should the system become overloaded, photon events may be lost and measurement times and display rates may become irregular.

You can minimize the running MultiHarp software during measurements without issues. This may be of interest for lengthy measurements in integration mode, where one is only interested in accumulating a certain amount of counts without need for permanent monitoring. However, the things you do in the meantime must not overload the CPU.

The "panel meters" showing the current count rates are not meant to be 100 % accurate. They merely serve as an aid for setting up the system. Some of them may suffer from system overload. Accordingly the values shown in the curve details (trace mapping dialog) are subject to such tolerances.

In TTTR mode, system overload will manifest itself in loss of data and termination of the measurement. Especially the real-time correlator requires significant resources. Here no other heavy load background processes should be allowed to run. The faster the PC, the less these issues will matter. TTTR mode performance (i.e. max. time tagging throughput to disk) may also be improved by using modern solid state disks.

7.2. Histogram Artefacts

Disturbing histogram ripple is strongly dependent on the quality of the input signals. Try to deliver the best possible signal quality with clean and reasonably steep active edges and without too much ringing. It is recommended to use detector and sync signals of similar amplitude to minimize crosstalk. If a picosecond pulsed laser from PicoQuant's PDL Series is used, attenuating the sync pulses by 10 dB immediately at the laser driver may reduce histogram ripple (set trigger level accordingly). Always use good quality 50 Ω components and coax leads with proper shielding and correct termination. We recommend cables of type RG223 with double braided and silver plated screen. Check your setup for ground loops. Grounding different system components (PC, detector, detector power supply, diode laser driver, etc.) at different points can induce considerable noise in the ground lines. Because signal return paths may share the same ground lines, this noise is copied into the signal lines and causes increased timing jitter and / or histogram ripple. Network cables and mobile phones may also be sources of noise. Use properly impedance matched power splitters (reflection-free T-pads) if signals must be fed to multiple 50 Ω inputs. Never use ordinary BNC Tees. All accessories are available from PicoQuant. PMT detectors should be connected through a suitable high speed preamplifier (available from PicoQuant). MCP-PMT detectors should be connected through an amplifier with slightly higher gain (also available from PicoQuant). TTL-SPAD-detectors (e.g. Perkin-Elmer SPCM-AQR and successors by Excelitas) must be connected through an attenuator or attenuating pulse inverter (e.g., PicoQuant SIA 400).

An often overlooked problem is pile-up and dead-time related histogram distortion. It becomes noticeable if the detector count rate exceeds ~5 % of the sync rate. This is why high excitation rates are so important. The effect is an inherent problem of high resolution TCSPC and not a fault of the hardware. The MultiHarp 150 is less significantly affected by such issues because of its very short dead-time. The safe way to handle dead-time is to maintain count rates < 5 % of the sync rate. Pulse pile-up may also be corrected for in data analysis. Indeed, combined with suitable detectors (e.g., PMA Hybrid from PicoQuant) it will allow count rates as high as 80 MHz, i.e. as fast as the excitation rate. For further details see the literature given at the end of section 2.4.

7.3. Warming Up Period

Observe the warming-up period of at about 20 minutes (depending on ambient temperature) before using the MultiHarp for important measurements. You can use this time for set-up and preliminary measurements. The maximum permissible ambient temperature is 35 °C. Make sure that the cooling air can circulate freely and no other hot instrument is directly under the MultiHarp 150.

7.4. Custom Programming of the MultiHarp

A programmer's library (DLL) for custom Windows software development is available to build your own applications e.g., in LabVIEW, Matlab, Python, C/C++, C#, and Pascal (Delphi/Lazarus). A rich set of demo code is provided for an easy start. If you care about performance, consider using a proper compiled high level programming language such as C/C++ or Pascal. There is also a library version for Linux (Intel processor architecture only) which is fully compatible with that for Windows so that applications can easily be ported across the two platforms.

7.5. Software Updates

We constantly improve and update the software for our instruments. This includes updates of the configurable hardware (FPGA). Such updates are important as they may affect reliability and interoperability with other products. The software updates are free of charge, unless major new functionality is added. The latest software is available for download at the PicoQuant web site. Alternatively you can click the button 'Check for Updates' in the Help-About dialog available through the main menu or the toolbar. Note that downloading a software update by means of this button does not automatically install it. Typically the downloads contain zip files that must be unpacked and installed manually. Also note that it may not always be advisable to blindly install the latest software. This is the case especially when a user or his/her team have developed custom software that relies on a certain file format. Updates may break this compatibility. Therefore, always carefully read the release notes before installing updates.

7.6. Support and Bug Reports

The MultiHarp 150 TCSPC system has gone through extensive testing. It builds on over 20 years of experience with several predecessor models and the feedback of hundreds of users. Nevertheless, it is a new product and some bugs may still be found. In any case we would like to offer you our support if you experience problems with the system. Do not hesitate to contact PicoQuant in case of difficulties with your MultiHarp.

As a first step it is always advisable to study the manual or to press F1 for help. Should you observe errors or bugs caused by the MultiHarp system please try to find a reproducible error situation. Then open *Help - Request Support*. This dialog provides important version information on the MultiHarp software and hardware (the latter only if it is connected and operational). It can be reached via the Help menu or via the toolbar. It provides a small text viewer window with a listing of MultiHarp hardware and software versions that you can copy and paste into your support request. Note that this information is very important for adequate troubleshooting. Support requests without this information cannot be processed and will be delayed by return questions for this information. If your system is not functional at all, the minimum information you must provide for support is the serial number of your MultiHarp. It can be found at the back of the housing. There may be other relevant circumstances, especially other new hardware installed in your PC, so please provide details. You can run *msinfo32* to obtain a listing of your PC configuration and attach the summary file to your error report to www.picoquant.com/contact/support. If you cannot access the web form directly, please contact support@picoquant.com and include the same information. Complete information will help us to help you more quickly. When you submit an error report referring to measurement data you have taken, attach the original data file. If the file is too large for email (>5 MB) please provide access to it on a public file server.

If the device must to be sent in for inspection / repair / upgrade, please request an RMA number before shipping the hardware. Observe precautions against static discharge, moisture, and mechanical damage under all circumstances in handling, packaging, and shipping. Use original or equally protective packaging material.

Of course we also appreciate good news: If you have obtained exciting results with one of our systems, please let us know, and where appropriate, please mention the instrument in your publications. At our web-site we maintain a large bibliography of publications related to our instruments. It may serve as a reference for you and other potential users. See <http://www.picoquant.com/scientific/references>. Please submit your publications for addition to this list.

8. Appendix

8.1. Warnings

When the MultiHarp software is running with functional hardware it continuously collects information about the input signals and the current acquisition settings. If these settings along with the input rates indicate possible errors, the software will indicate this by showing a warning icon.



While the software is running in interactive histogramming mode the warning icon is displayed at the bottom of the main window in the status bar. In the case of TTTR mode, it will appear directly in the TTTR mode dialog. The icon can be clicked to display a list of current warnings together with a brief explanation of each warning. Similarly the Warnings (if any) will be stored in the data files and can be inspected via the Curve Details dialog, also after re-loading such a file later.

The warnings are to some extent dependent on the current measurement mode. Not all warnings will occur in all measurement modes. Also, count rate limits triggering a specific warning may be different in the various modes. The following table lists the possible warnings in the three measurement modes and gives some explanation as to their possible cause and consequences.

| Warning | Histo Mode | T2 Mode | T3 Mode |
|--|------------|---------|---------|
| WARNING_SYNC_RATE_ZERO No pulses are detected at the sync input. In histogramming and T3 mode this is crucial and the measurement will not work without this signal. | √ | | √ |
| WARNING_SYNC_RATE_VERY_LOW The detected pulse rate at the sync input is below 100 Hz and cannot be determined accurately. Other warnings may not be reliable under this condition. | √ | | √ |
| WARNING_SYNC_RATE_TOO_HIGH The pulse rate at the sync input (after the divider) is higher than 75 MHz. This is close to the TDC limit. Sync events will be lost above 78 MHz. T2 mode is normally intended to be used without a fast sync signal and without a divider. If you see this warning in T2 mode you may accidentally have connected a fast laser sync. | √ | √ | √ |
| WARNING_INPT_RATE_ZERO No counts are detected at any of the input channels. In histogramming and T3 mode these are the photon event channels and the measurement will yield nothing. You might sporadically see this warning if your detector has a very low dark count rate and is blocked e.g. by a shutter. In that case you may want to disable this warning. | √ | √ | √ |
| WARNING_INPT_RATE_TOO_HIGH The overall pulse rate at the input channels is higher than 80 MHz (USB 3.0 connection) or higher than 9 MHz (USB 2.0 connection). This is close to the throughput limit of the present USB connection. The measurement will likely lead to a FIFO overrun. There are some rare measurement scenarios where this condition is expected and the warning can be disabled. Examples are measurements where the FIFO can absorb all data of interest before it overflows. | √ | √ | √ |

| Warning | Histo Mode | T2 Mode | T3 Mode |
|--|------------|---------|---------|
| WARNING_INPT_RATE_RATIO This warning is issued in histogramming and T3 mode when the rate at any input channel is higher than 5% of the sync rate. This is the classic pile-up criterion. There are some measurement scenarios where this condition is expected and the warning can be disabled. Examples are antibunching measurements or Rapid-FLIM where pile-up is either tolerated or corrected for during data analysis. One can usually also ignore this warning when the current time bin width is larger than the dead-time. | √ | | √ |
| WARNING_DIVIDER_GREATER_ONE In T2 mode: The sync divider is set larger than 1. This is probably not intended. The sync divider is designed primarily for high sync rates from lasers and requires a fixed pulse rate at the sync input. In that case you should use T3 mode. If the signal at the sync input is from a photon detector (coincidence correlation etc.) a divider > 1 will lead to unexpected results. There are rare measurement scenarios where this condition is intentional and the warning can be disabled. In histogramming and T3 mode: If the pulse rate at the sync input is below 75 MHz then a SyncDivider >1 is not needed. The measurement may yield unnecessary jitter if the sync source is not very stable. | √ | √ | √ |
| WARNING_DIVIDER_TOO_SMALL The pulse rate at the sync input (after the divider) is higher than 75 MHz. This is close to the TDC limit. Sync events will be lost above 78 MHz. To avoid this, increase the sync divider. | √ | | √ |
| WARNING_TIME_SPAN_TOO_SMALL This warning is issued in histogramming and T3 mode when the sync period (1/SyncRate) is longer than the start to stop time span that can be covered by the histogram or by the T3 mode records. You can calculate this time span as follows: $\text{Span} = \text{Resolution} * \text{Length}$ Length is 32768 in T3 mode. In histogramming mode it depends on the chosen histogram length (default is 65536). Events outside this span will not be recorded. There are some measurement scenarios where this condition is intentional and the warning can be disabled. | √ | | √ |
| WARNING_OFFSET_UNNECESSARY This warning is issued in histogramming and T3 mode when an offset >0 is set even though the sync period (1/SyncRate) can be covered by the measurement time span (see calculation above) without using an offset. The offset may lead to events getting discarded. There are some measurement scenarios where this condition is intentional and the warning can be disabled. | √ | | √ |
| WARNING_COUNTS_DROPPED This warning is issued when the front end of the data processing pipeline was not able to process all events that came in. This will occur typically only at very high count rates during intense bursts of events. | √ | √ | √ |

If any of the warnings you receive indicate wrong pulse rates, the cause may be inappropriate input settings, wrong pulse polarities, poor pulse shapes or bad connections. When in doubt, check all signals with an oscilloscope of sufficient bandwidth.

Note that the software can detect only a subset of all possible problematic measurement conditions. It is therefore not safe to assume "all is right" just by seeing no warning. On the other hand, if any of the warnings turns out to be an unnecessary nuisance, e.g., because your specific measurement conditions inevitably trigger it, you can disable that warning via the General Settings dialog (see section 6.7). Note, however, that disabling the warnings will also apply to which warnings you get to see in the Curve Details dialog when an existing data file is inspected. It may be better then to enable all warnings.

8.2. Data File Formats

While for many purposes the ASCII export of histograms to files or to the clipboard is sufficient and easy, you also may want to access the MultiHarp data files via custom programs. This section provides only a brief overview on the file format. For details please refer to the online help file available via the help menu.

To overcome certain limitations of various different formats used in the past, PicoQuant now uses a unified file format. It is designed to be future proof in the sense that files created by a current software version stay valid for future software revisions and, moreover, files created by future software versions will most likely still be readable by older software, although they might contain information, that software can't even "know" about. This is achieved by using a tagged format. Tags identify the data to follow, and give the type, length and even meta information. The exact location of an individual item in the file is then irrelevant. Version robustness is granted as long as version-breaking changes to the semantics of a given field are implemented by a tag with a new identifier rather than expanding the range or interpretation of the old one. The list of tags (identifiers) and their interpretation rules can be kept in a tag dictionary. With this as a precondition, the software only has to show tolerance on missing non-mandatory (i.e. optional) content.

The new format definition unifies PicoQuant's existing file formats which individually evolved over many years. The resulting new TTTR file format with the extension `*.ptu` will be used for all current and future TCSPC products supporting TTTR mode and enriches them with powerful new features. Similarly, a tagged file format with the extension `*.phu` covers the histogram data formats of our current and future TCSPC products.

To support understanding of the format and implementation of custom software accessing these files, a set of demos is provided in the subfolder `\Filedemo` in your chosen software installation folder. If you need to evaluate more header items than the demos do, please refer to the MultiHarp online help file available via the help menu. A file format related HTML help file is also provided in the file demo folder. It contains a list of tag types and a tag dictionary that explains the individual items. Note that the dictionary contains more items than the MultiHarp software actually uses. It is recommended to go by a specific file, have one of the demos read it and then look at the list of header items you get. You can also use the PicoQuant File Info shell extension that will be installed by the MultiHarp software setup to inspect individual header items of a `*.ptu` or `*.phu` file. Just right-click on the file in Windows explorer and select *Properties*.

Despite the intended version tolerance of the tagged format, for consistency and safe version checking the MultiHarp data files still carry a format version number, which is now called *content version* and currently has the string value "1.0". In order to identify a MultiHarp data file as a file created by and to be used by the native MultiHarp software there is a tag *assured content* which begins with the string "MultiHarp". There is also a pair of tags creator name and creator version that identify the creating software. Programmers of custom software writing such files MUST USE THEIR OWN CREATOR NAME.

Note that despite our best efforts towards compatibility and version tolerance, file formats in future software releases are subject to change without notice.

8.2.1. Interactive Mode File Format

The standard MultiHarp histogram data files created by the MultiHarp software in interactive histogramming mode (`*.phu`) are tagged binary files which contain both the setup parameters and the actual histogram data. The latter can be present multiple times, i.e. multiple measurements can be stored in one file. Relevant settings are stored for each measurement separately. In order to identify a MultiHarp data file as a file created by and to be used by the native MultiHarp software, a program reading in these files can read the tag *assured content* which begins with the string "MultiHarp". However, a piece of software aiming solely at retrieving the histogram data content can (and should) be tolerant about this tag and go for the pure histogram data. This tolerance will ensure compatibility for the future. Indeed, the demos in the subfolder `\Filedemo` in your chosen installation directory are following this tolerant approach. For more information on individual file tags and their content, please consult the online help file available via the help menu.

8.2.2. TTTR Mode File Format

MultiHarp data files from T2 and T3 Mode (`*.ptu`) created by the MultiHarp software are tagged binary files which contain both the setup parameters and the actual event data. There can be only one measurement per file. The setup data in the file header is similar to that in standard interactive mode files. In order to identify a MultiHarp data file as a file created by and to be used by the native MultiHarp software, a program reading in these files can read the tag *assured content* which begins with the string "MultiHarp". However, a piece of soft-

ware aiming solely at retrieving the event record data content can (and should be) be tolerant about this tag and go for the pure event record data. This tolerance will ensure compatibility for the future. Indeed, the demos in the subfolder `\Filedemo` in your chosen installation directory are following this tolerant approach. For more information on individual file tags and their content, please consult the online help file available via the help menu. It is worth noting that the actual TTTR record data following the file header corresponds directly to the raw data obtained with custom programs using the MultiHarp programming library.

8.3. Hardware Technical Data

8.3.1. Specifications

All information given here is reliable to our best knowledge. However, no responsibility is assumed for possible inaccuracies or omissions. Specifications and external appearance are subject to change without notice. The MultiHarp 150 can be ordered with 4, 8, or 16 input channels. Note that more channels cannot later be added.

Input Channels and Sync

| | |
|---|--|
| Trigger principle | edge trigger software adjustable |
| Trigger edge | rising or falling software adjustable |
| Input impedance | 50 Ω |
| Input voltage operating range (pulse peak) | -1200 ... 1200 mV |
| optimum: | abs. Amplitude 150..300 mV |
| Input voltage max. range (damage level) | ± 2500 mV |
| Trigger level adjust | -1200 ... 1200 mV |
| Input pulse width | ≥ 0.4 ns |
| Input pulse rise/fall time | ≤ 20 ns |
| Marker and control inputs | standard TTL, 100 k Ω DC load |

Time to Digital Converters

| | |
|--|---|
| Base resolution (min. time bin width) | 80 ps |
| Single measurement timing uncertainty (rms)* | < 60 ps |
| Start-stop timing uncertainty (rms)* | $< 85 \text{ ps} + 10^{-8} \cdot \Delta t$ |
| Differential non-linearity | < 10% p.p. < 1% rms |
| Dead time | < 650 ps |
| Peak count rate | $1.5 \cdot 10^9$ cps sustainable for bursts of 2000 events |
| Max. sustained count rate | $78 \cdot 10^6$ cps |
| Max. sync rate without divider | 78 MHz |
| Max. sync rate with divider | 1200 MHz |
| Adjustable delay range for each input | ± 100 ns |
| delay resolution | 80 ps |

* In order to determine the timing uncertainty or "jitter" it is necessary to repeatedly measure a time difference and to calculate the standard deviation (rms error) of these measurements. This is done by splitting an electrical signal from a pulse generator and feeding the two signals each to a separate input channel. The differences of the measured pulse arrival times are then calculated along with the corresponding standard deviation. This latter value is the rms jitter or timing uncertainty of start-stop measurements. However, calculating such time differences requires two measurements. These both have a small random jitter and a term that is dependent on the time difference between the two signals because over longer spans the crystal clock jitter becomes noticeable. For short term measurements on a sub-microsecond scale, as in most TCSPC applications, the latter is typically negligible. The timing precision of a single measurement, according to error propagation laws, is obtained by dividing the start-stop timing uncertainty by $\sqrt{2}$. We specify this single measurement rms error here for comparison with other products.

Histogrammer

| | |
|--|---|
| Maximum number of time bins | 65536 |
| Full scale time range | 5.24 μ s to 21.99 s (depending on chosen resolution) |
| Count depth per time bin | 4,294,967,296 (32 bit) |
| Acquisition time | 1 ms ... 100 h |
| Sustained throughput to device memory (sum of all channels) | 180 · 10 ⁶ cps |

TTTR Engine

| | |
|---|--|
| T2 mode resolution | 80 ps |
| T3 mode resolution (settable) | 80 ps, 160 ps, 320 ps, ..., 2.68 ms |
| T3 Mode number of time bins | 32768 |
| FIFO buffer depth (records) | 134,217,728 |
| Sustained throughput (sum of all channels) | typ. 80 × 10 ⁶ events/sec (depending on host PC configuration and performance) |
| Acquisition time | 1 ms ... 100 h |
| External markers (TTL in) | 4 |
| External marker input pulse duration | > 50 ns |
| External marker input pulse spacing | > 50 ns |
| External marker rise/fall time | < 50 ns |
| External Marker timing error in T2 mode | < 100 ns |
| External Marker timing error in T3 mode | < Tsync + 100 ns |

External Reference Clock

| | |
|--------|---|
| Input | 10 MHz 200 ... 1 500 mV p.p. 50 Ω ; AC coupled |
| Output | 10 MHz 1400 mV p.p. 50 Ω ; AC coupled |

Trigger Output

| | |
|---------------------------|-------------------------|
| Baseline level | 0V typ. |
| Pulse amplitude | -0.7V typ. |
| Pulse duration | 10 ns typ. |
| Load/Termination | 50 Ω |
| Programmable period range | 0.1 μ s ... 1.678 s |

Operating Environment

| | |
|------------------------------|------------------------------|
| Ambient temperature | $\leq 35^{\circ}\text{C}$ |
| Warm-up period to meet specs | 20 min |
| Recommended PC specs | ≥ 2 cores, ≥ 2 GHz |
| CPU | ≥ 4 GB |
| RAM | USB 2.0 |
| USB minimum required | USB 3.0 or USB 3.1 |
| USB required to meet specs | |
| Operating system | Windows 8 or 10 |

Power Consumption

| | |
|-------------------------|---------------------------|
| Supply Voltage | 100...240V AC, 50...60 Hz |
| Total power consumption | ≤ 50 W |

Dimensions

| | | |
|-------------------------|------------------------|------------------|
| MultiHarp 150 4N and 8N | incl. feet and handles | 305× 240 × 95 mm |
| MultiHarp 150 16N | incl. feet and handles | 305× 350 × 95 mm |

Retraction of Discarded Devices

Waste electrical products must not be disposed of with household waste.

This equipment should be taken to your local recycling centre for safe treatment.



WEEE–Reg.–Nr. DE 96457402

8.3.2. Connectors

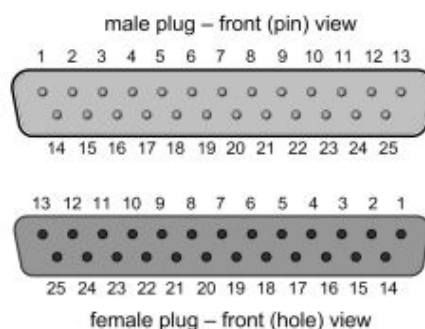
The inputs for the photon detector signals and the sync signal are SMA connectors located on the front panel of the MultiHarp 150. They are labelled SYNC, CH1..CH4 (or CH1..CH8 depending on the model). The inputs are terminated with 50 Ohms internally. Use quality 50 Ohms coax cables with appropriate connectors. For interfacing to BNC connectors use standard adapters. Carefully screw on the SMA connectors for sync and detector until they are hand-tight. Do not use wrenches. Note that PMT detectors operate with high voltages that may discharge through the signal cable. Make sure such detectors are switched off and fully discharged before connecting them.

Apart from the SMA connectors for the input signals the MultiHarp has further connectors for USB, White Rabbit, clock synchronization, and experiment control.

The USB connector (blue) is a standard USB 3.0 type B socket located at the front panel. It is used to connect to the host PC via a standard USB 3.0 cable. Observe the notes in section 3.4 for making this connection.

The White Rabbit (WR) connector, also on the front panel, is designed to hold exchangeable SFP transceiver modules. Upon shipping such a module is not installed. Suitable SFP transceiver modules can be ordered from PicoQuant as well as from other vendors.

The control connector is a 25-pin female sub-D connector labeled 'CTRL'. In case of the MultiHarp 150 4N and 8N it is located on the front panel and in case of the MultiHarp 150 16N it is located at the back of the housing. The following figure shows the pin layout and the table below contains the connector's default pin assignments. Note that future firmware/software is going to allow reconfiguration of some of these pin assignments.



CTRL Connector – Pin numbering scheme

| CTRL Connector – Default Pin Assignments | | | |
|--|---------|---------|---|
| Pin# | Name | I/O | Purpose/Description |
| 1 | GPIO 0 | TTL in | marker 1 input |
| 2 | GPIO 1 | TTL in | marker 2 input |
| 3 | GPIO 2 | TTL in | marker 3 input |
| 4 | GPIO 3 | TTL in | marker 4 input |
| 5 | GPIO 4 | | reserved |
| 6 | GPIO 5 | | reserved |
| 7 | GPIO 6 | | reserved |
| 8 | GPIO 7 | | reserved |
| 9 | GPIO 8 | | reserved |
| 10 | GPIO 9 | | reserved |
| 11 | GPIO 10 | | reserved |
| 12 | GPIO 11 | | reserved |
| 13 | GPIO 12 | | reserved |
| 14 | GND | GND | Ground (0V) |
| 15 | GPIO 13 | | reserved |
| 16 | GPIO 14 | | reserved |
| 17 | GPIO 15 | TTL in | GPS RS232 RX (3.3V signal level) for connection to GPS receiver |
| 18 | GND | GND | ground (0V) |
| 19 | C1 | TTL in | start measurement (requires dedicated software) |
| 20 | C2 | TTL in | stop measurement (requires dedicated software) |
| 21 | GND | GND | ground (0V) |
| 22 | MACT | TTL out | high when measurement running |
| 23 | GND | GND | ground (0V) |
| 24 | D3V3 | DC out | +3.3V / ≤ 350 mA supply for external hardware add-ons |
| 25 | GND | GND | ground (0V) |

The specifications for TTL signals can be found at http://wikipedia.org/wiki/Transistor-transistor_logic.

For further details regarding signal specifications, notably the trigger output (voltages, frequencies pulse widths etc.) see section 8.3.1.

Pins 1, 2, 3 and 4 accept TTL compatible synchronization signals that will be recorded as markers in TTTR mode. The pins are internally pulled down, so that they are inactive when left unconnected. The active edge is chosen in the software settings dialog. Rise/fall times must be 50 ns or faster. Both high and low state must be at least 50 ns long. The clock period may therefore (in principle) be as short as about 100 ns but data bus throughput constraints will apply. Each marker creates an additional TTTR record, so that one must ensure not to swamp the data stream with too many marker records. When bandwidth gets tight, markers take precedence over photon records, so that excess marker traffic can suppress photon records. In fast imaging applications it is therefore recommended not to use a pixel clock but a line clock only. Because each photon has a time tag, it is usually not necessary to use an additional pixel clock. For more information on how to use the marker inputs see section Fehler: Verweis nicht gefunden.

Pin 17 can be used to connect the serial TX line of a GPS receiver for transmission of GPS time.

Pins 19 and 20 can be used to implement hardware triggered measurements. Note that this requires custom software (see the DLL manual and related demos).

Pin 22 is a TTL output that goes high when a measurement is running.

Pin 24 provides a 3.3 V DC supply voltage that external electronics can use. Under no circumstances must this line be shorted to ground or loaded in excess of the specified maximum current.

Pins 14, 18, 21, 23, and 25 are the common ground for the TTL signals and the DC supply line. They are also connected to the housing.

Make sure not to confuse the control connector with a connector for other equipment that would physically fit but might lead to mutual damage. Appropriate cables for typical applications of the control port are available from PicoQuant.

At the back of the housing the MultiHarp 150 provides a standard connector for power supply and another four SMA connectors labelled "TRG OUT", "REF IN", "REF OUT", "PPS IN".

TRG OUT is a programmable trigger output for pulsed light sources. Note that this is under software control and must be used with greatest care when potentially dangerous laser sources are triggered from it. The trigger rate is programmable from 0.1 μ s to 1.678 s (0.596 Hz to 10 MHz). The pulse width is about 10 ns, the base line level is near 0 V and the active level (pulse peak) is approximately -0.7 V. When a light source is triggered from this signal the same signal must typically be fed to the sync input. This requires proper impedance matching e.g., by means of a power splitter (reflection-free T-pad).

REF IN accepts an industry standard 10 MHz clock reference, e.g., from a frequency normal, an atomic clock, or another MultiHarp. When this clock source is selected (via software) then the MultiHarp will lock its internal PLL to this signal. Note that it must not be disconnected once it is being used as the active clock source.

REF OUT provides an industry standard 10 MHz clock output phase locked to the MultiHarp's internal clock, e.g., for the REF IN of another MultiHarp 150. Once the MultiHarp has locked its internal PLL to this signal the clocks of the two devices will not drift apart.

PPS IN accepts the "pulse per second" signal of e.g., GPS receivers. In combination with the 10 MHz clock and the time code transmitted by the GPS receiver it is possible to synchronize the MultiHarp with clock and time from GPS. This can be used to synchronize two or more MultiHarp to GPS and one another. The accuracy of such a synchronization depends on that of GPS and the quality of the receiver. It is typically on the order of some tens of ns. The benefit is remote synchronization over large distances. If better accuracy is required, consider White Rabbit instead.

8.3.3. Indicators

The MultiHarp shows some status information by means of three LEDs on the front panel labelled STATUS. The meaning of these indicators, from left to right, is as follows:

USB Status (left)

red = no connection
orange = USB 2.0 connection
green = USB 3.0 connection

Measurement Status (middle)

red = measurement active
green = inactive

This reflects the state of pin 22 of the CTRL connector (high when measurement running).

Error Status (right)

red = error (or not yet initialized)
orange = info
green = OK

Note that the error status LED is initially red when the device is switched on and the software has not yet initialized it. Also note that it gives only a very crude indication of errors. The indicated errors or info states are also monitored by the software which gives much more tangible information on the issue. The most likely info you may see indicated by orange light is when the input rate is too high and events are dropped. However, espe-

cially in case of infos the LED may actually light up only very briefly. Looking at the status information given by the software should therefore always be preferred.

8.4. Using the Software under Linux

The MultiHarp software can also be used under Linux (x86 platform only). This requires that Wine is installed (see <https://www.winehq.org>). You can run the regular software setup as explained in section 3.3. Instead of installing a device driver, running under Linux with Wine requires that you have Libusb 1.0 installed (see <https://libusb.info/>). We have successfully tested with Wine 3.0 and Libusb 1.0.19 on Linux Mint 19.1.

Libusb Access Permissions

For device access through libusb, your kernel needs support for the USB filesystem (usbfs) and that filesystem must be mounted. This is done automatically, if `/etc/fstab` contains a line like this:

```
usbfs /proc/bus/usb usbfs defaults 0 0
```

This should routinely be the case if you installed any of the mainstream Linux distributions.

The permissions for the device files used by libusb must be adjusted for user access. Otherwise only root can use the device(s). The most appropriate way of setting the suitable permissions is by means of hotplugging scripts or udev. Which mechanism you can use depends on the Linux distribution you have. Most recent distributions use udev.

Udev

For automated setting of the device file permissions with udev you have to add an entry to the set of rules files that are contained in `/etc/udev/rules.d`. Udev processes these files in alphabetical order. The default file is usually called `50-udev.rules`. Don't change this file as it could be overwritten when you upgrade udev. Instead, put your custom rule for the MultiHarp in a separate file. The contents of this file should be:

```
ATTR{idVendor}=="0d0e", ATTR{idProduct}=="0013", MODE="666"
```

A suitable rules file `MultiHarp150.rules` is provided in the folder `Linux/udev` on the distribution media. You can simply copy it to the `/etc/udev/rules.d` folder. The install script in the same distribution media folder does just this. Note that the name of the rules file is important. Each time a device is detected by the udev system, the files are read in alphabetical order, line by line, until a match is found. Note that different distributions may use different rule file names for various categories. For instance, Ubuntu organizes the rules into further files: `20-names.rules`, `40-permissions.rules`, and `60-symlinks.rules`. In Fedora they are not separated by those categories, as you can see by studying `50-udev.rules`. Instead of editing the existing files, it is therefore usually recommended to put all of your modifications in a separate file like `10-udev.rules` or `10-local.rules`. The low number at the beginning of the file name ensures it will be processed before the default file. However, later rules that are more general (applying to a whole class of devices) may later override the desired access rights. This is the case for USB devices handled through Libusb. It is therefore important that you use a rules file for the MultiHarp that gets evaluated after the general case. The default naming `MultiHarp150.rules` most likely ensures this but if you see problems you may want to check.

Note that there are different udev implementations with different behavior. On most recent Linux distributions it is sufficient to just re-plug the device after installing the rules file. On some distributions you must reboot to activate changes, on others you must reload the rules and/or restart udev.

Wine Limitations

A limitation is the current implementation of Wine's viewer for chm help files. You may observe failure or some glitches when using the online help facility. Another issue is that when running the MultiHarp software under Linux with Wine the WhiteRabbit configuration does not work properly.

Finally, please note that running the MultiHarp software under Linux with Wine is an experimental feature that cannot be covered by regular product support.

All information given in this manual is reliable to our best knowledge. However, no responsibility is assumed for possible inaccuracies or omissions. Specifications and external appearance are subject to change without notice.



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