PUMA Foundation Scope

User's Manual

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Manual prepared by **OptArc** – Designers and manufacturers of custom optical and optoelectronic systems using 3D printing rapid prototyping and based in the UK.
See **optarc.co.uk** for details.

Safety Information

Identification of risk

Throughout this manual please take heed of **warnings given in bold text and highlighted yellow** to avoid possible damage to equipment and/or harm to people.

Risk to vulnerable groups

PUMA microscopes and associated systems are not toys. They contain small parts which may come loose such as tiny metal screws and washers and glass components that may splinter or break or otherwise present a choking or sharp object hazard or chemical hazard (e.g. for batteries). **Please do not let babies or young children play with or gain access to any aspect of a PUMA system without close appropriate adult supervision. Likewise keep PUMA systems away from pets**.

Risk of damage to eyesight

When choosing and using a light source for a PUMA microscope, care must be taken to avoid the use of or exposure to light that could damage eyesight.

In particular **never allow direct or specular reflections of the sun to enter the microscope** through the illuminating mirror.

Also, **never allow laser light to enter the microscope through the illuminating mirror if any viewing is to be done by eye** (as opposed to recordings by a camera only).

Download link for PDF version of this Manual

You can download the latest version of this User's Manual as a PDF file from the Support section of the OptArc website via this link:

https://www.optarc.co.uk/support/

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Legal Information

Copyright

Trade Marks

OptArc and the OptArc logo are Registered Trade Marks of Dr Paul J. Tadrous (registered with the UK Intellectual Property Office).

Limitations of Use

The PUMA microscope and its associated systems do not have any certifications or regulatory approvals in any country for use in clinical diagnostics or treatment (human or veterinary).

The PUMA microscope and its associated systems are released to be used for research and educational purposes only.

Disclaimer

All PUMA project information, including without limitation any CAD file or STL file and all documentation, advice and instruction (whether provided in video form, audible form, written form or otherwise) is provided ‘as is’ in good faith and is intended to be helpful but comes with no warranty whatsoever.

Anyone attempting to build or use a PUMA microscope or other PUMA-related material, accessory, module or derivative is hereby advised that there will be risk involved in 3D printing, post-print processing, assembly and usage of the resulting structures. This risk includes, without limitation, the risk of personal damage and loss of resources.

Dr Paul J. Tadrous, TadPath and OptArc cannot accept any liability for any such loss or damages that may occur. All those who attempt to build or use any aspect of the PUMA project or derivatives thereof do so at their own risk.

Discrepancy in Appearance of Parts

The parts in your package may differ in exact appearance to the parts shown in this manual or in the associated videos or advertising materials because we always ship the latest versions of the scope and the manual and videos may have been prepared using earlier models. Functionality of the parts you receive will be the same or better than those illustrated in this manual and any associated video or advertising materials.

Abbreviations

Some common abbreviations used in this manual are listed below for convenience.

AR	Augmented Reality
CAD	Computer Aided Design
HUD	Heads-up Display
IC	Integrated Circuit
LED	Light Emitting Diode
NA	Numerical Aperture
PLA	Poly-lactic acid
PUMA	Portable Upgradeable Modular Affordable
QR	Quick Release (mechanism for attaching the optical tube)
RMS	Royal Microscopical Society
SLM	Spatial Light Modulator
STL	Stereolithography file (a file format for meshes)

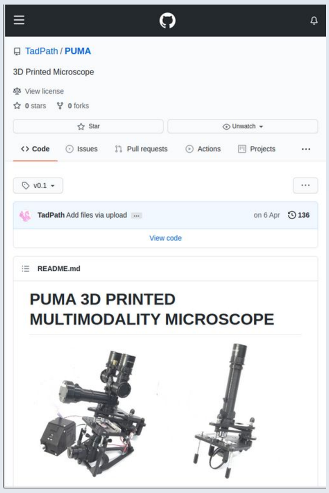
Introduction

PUMA

PUMA is an open source 3D printed high quality customisable microscope system. The name PUMA stands for ***P***ortable, ***U***pgradeable, ***M***odular and ***A***ffordable – key features of this system. To learn more about the PUMA open source project and to get CAD files, software and schematics please see the official PUMA GitHub repository:

PUMA GitHub Page

https://github.com/TadPath/PUMA



PUMA on GitHub

Foundation scope


Foundation scope

The ***Foundation scope*** is the simplest configuration of a PUMA system that can be used as a working microscope. This configuration is made available for sale by OptArc so that people who, for whatever reason, don't want to build a PUMA scope from scratch using DIY 3D printing can easily get started with PUMA. Anyone with 3D printing and DIY abilities could also make their own Foundation scope using the standard PUMA modules and specifications available on the GitHub page.

It is called 'foundation' because you can use this scope as a starting point from which to swap in various optional extra modules to build any of the other PUMA configurations all the way to an advanced research microscope with motorised Z-stage and TFT screens for augmented reality optical image overlay and Fourier optics digital light processing, binocular or trinocular viewports, fluorescence, phase contrast, polarisation, and more.

OptArc high quality x10 eyepiece

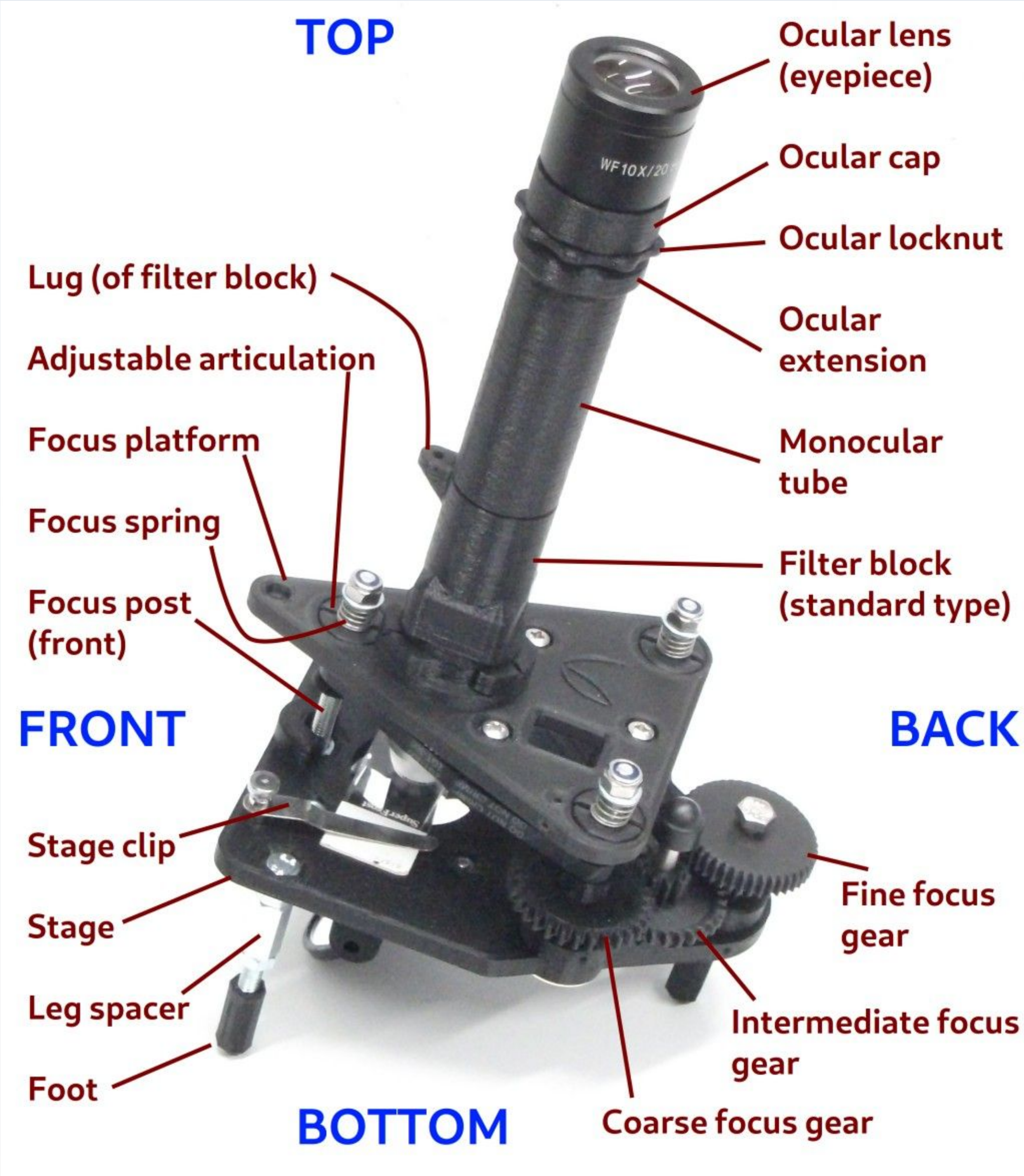
If purchased with the option to include optics, the OptArc PUMA Foundation scope comes with a high quality Plan Achromat objective and a x10 high quality ocular with good eye relief and a wide exit pupil for comfortable viewing. With its mirror illuminator alone the cope can give good images with x4 and x10 objectives (total magnification x40 or x100 respectively).

If you want to use even higher magnifications then you are advised to add the PUMA Abbe condenser (this is an optional upgrade). See the 'Upgrades' chapter later in this manual for more complete information about the various upgrades available.

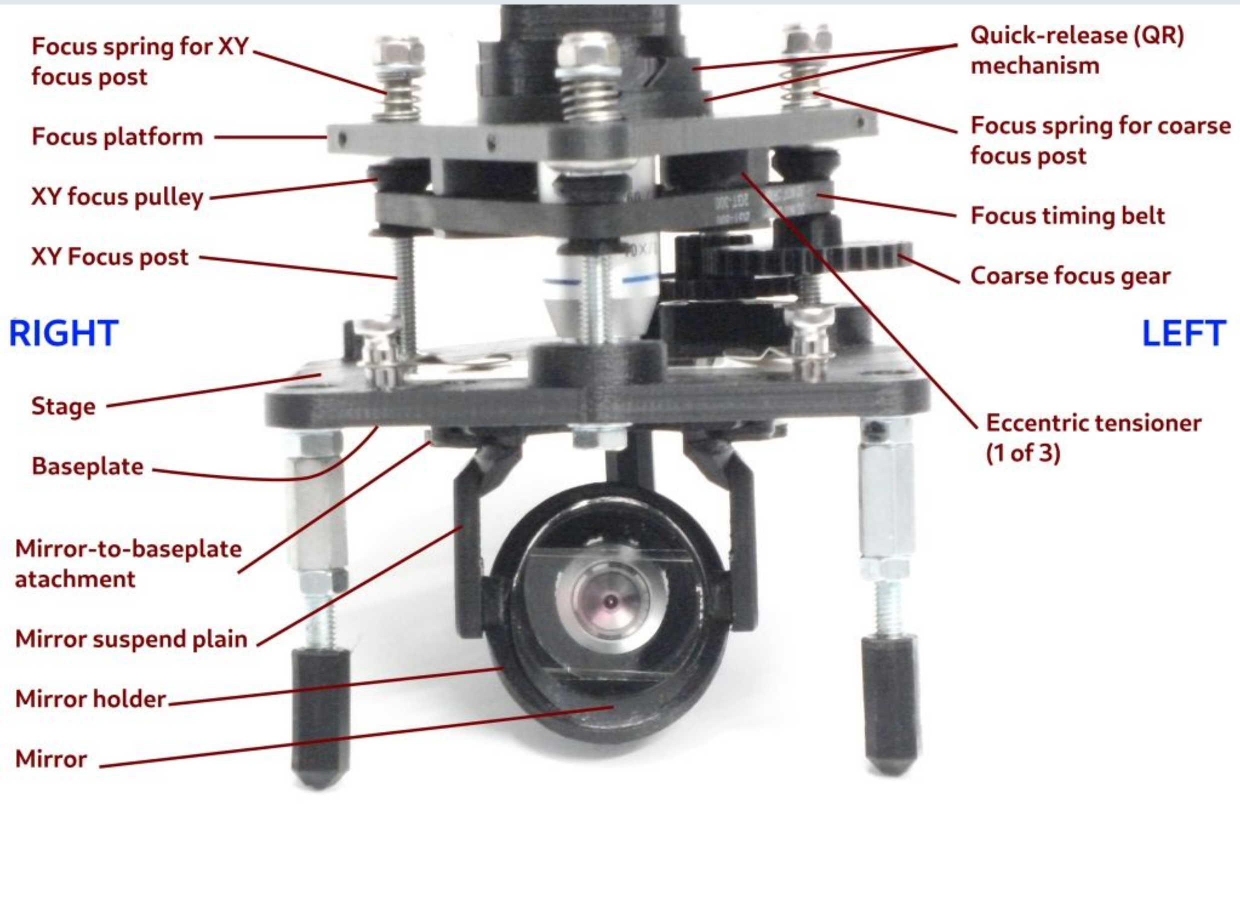
Anatomy of a PUMA Foundation Scope

The names of various parts of your Foundation scope are explained here. More detailed technical specs and nomenclature are available on the PUMA GitHub pages but this information should be all you need for everyday use of your scope and common maintenance as well as understanding the instructions in the rest of this manual.

Note that these illustrations may differ in aspects of detail from your Foundation scope because they were prepared with slightly earlier prototypes (for example the leg spacers contain an extra nut on them which is not part of the spec of a modern Foundation scope) but none of these differences invalidate the nomenclature.



The terms LEFT and RIGHT refer to the user's left and right when observing the microscope from behind (which is where the primary user is most commonly expected to be situated when making observations). This explains why those terms appear to be reversed in the front-on view shown in the figure below.



The terms 'stage' and 'baseplate' are interchangeable. Baseplate is used more commonly when referring to the undersurface of the stage. The term 'Z-stage' refers to the whole focussing mechanism i.e. the stage/baseplate and the focus platform as joined by the three focus post bolts with a timing belt and pulley mechanism (this arrangement is known as the 'tripost architecture' of the Z-stage).

The 'optical tube' refers to all above-stage optics plus the objective and this fits onto the focus platform via a tri-helical flexure thread called the 'quick release mechanism' or simply the 'QR mechanism'. This is composed of a male part (attached to the bottom of the filter block and which contains the thread for the objective) which mates with a female part that is an integral component of the focus platform.

The mirror illumination system is composed of several parts as shown. The mirror itself is the glass oval mirror. This is fixed to the oval plastic 'mirror holder' which, in turn, articulates with the 'mirror suspend plain' component. The latter component is held against the baseplate by the 'mirror-to-baseplate attachment' via two M4 thumbscrews (illustrated in the section 'Assembling the Microscope' below).

There are other features present which are not described here because they are only relevant to scopes that have upgrades fitted. These will be described in any manuals or tutorials pertaining to the fitting and use of those upgrade modules.

Unboxing and Initial Assembly

PUMA Unboxing Video

https://youtu.be/EfecD0UGLDQ

Unboxing

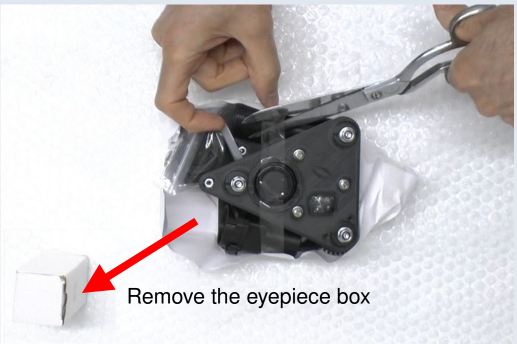
The following describes the recommended procedure for removing the contents of the box that your Foundation Scope shipped in. This procedure was designed to protect the sensitive optics. An unboxing YouTube video has been prepared to illustrate the following steps and can be accessed via the link shown. If you purchased a scope without eyepiece or objective then some of the following will not apply.

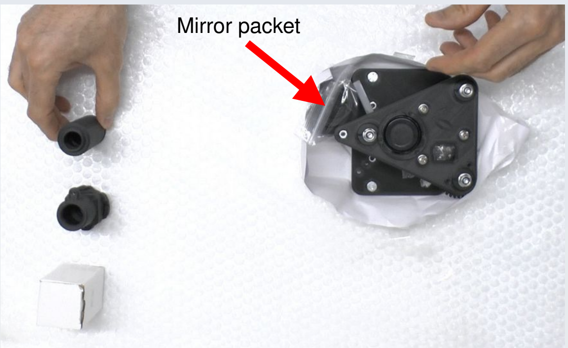
Step 1. Ensure the box is opened from the top. Take care not to damage the contents when opening (do **not** stick a knife in to cut open the box sealing tape).

Step 2. Remove the instruction manual and top loose packing filler material to reveal the bubble-wraped package which contains all the parts.

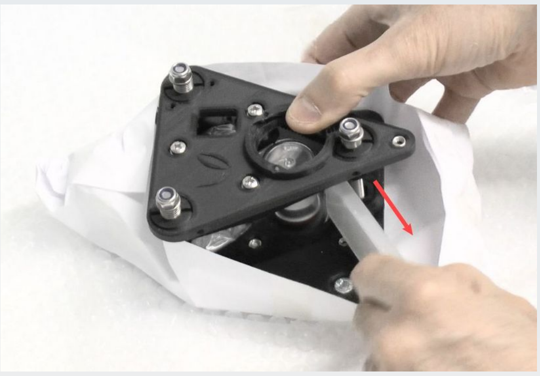
Step 3. Gently lift out the bubble-wrapped package from the box and lay it on a flat solid work surface. Remove the box and loose packaging filler material from the work area. The package is enclosed in two layers of bubble wrap. Cut the tape to release the first layer of bubble wrap. The parts will be seen – through the second, inner layer of bubble wrap – in a ‘boat’ made from paper.

Step 4. The paper boat is the bottom aspect. Ensure you keep the contents with this 'boat' side down while you cut the tape that holds the inner layer of bubble-wrap and remove it. **Keep everything on the work surface. from now until step 6 – do not try to hold it freely in the air or the objective lens may fall through the bottom and be damaged**. Remove the small white box that contains the ocular lens and put it to one side on the work surface. (as shown). Cut the tape that holds everything in place in the 'boat' and remove the simple filter block and the monocular tube and lay them to one side. Also remove the mirror packet and put it to one side.



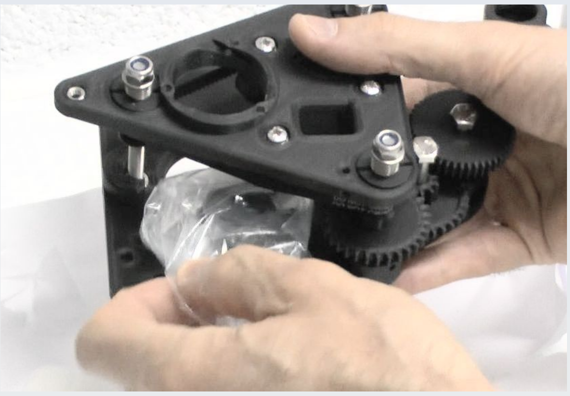


Step 5. Remove the test slide in its protective case by sliding it out from under the timing belt as shown.



Step 6. Tear the paper at the back of the paper boat and open out the paper but do not remove it from under the package of parts yet – keep it in situ. **Keep one hand over the top aperture as shown in the picture and slowly rotate the stage up so you can access the bottom aspect where the objective will come out. It is only held there by a piece of sticky tape so do not lift the scope way off the bench, just rotate it so you can see the underside and remove the protective cap from the undersurface and the objective after that** (see pictures below). You can now remove the protective cap that was taped across this bottom aperture and keep it to one side (this is a protective cap for your eyepiece).

Step 7. Remove the paper boat completely and remove the 'various small parts package' that is sandwiched in between the stage and focus platform near the back (as shown).



Step 8. Take the parts out of the 'various small parts package' and lay them out. Also remove the parts from the package containing the mirror – these will be the mirror with its holder plus the ocular lens holder assembly.

You are now ready to progress to assembling the microscope as described below.

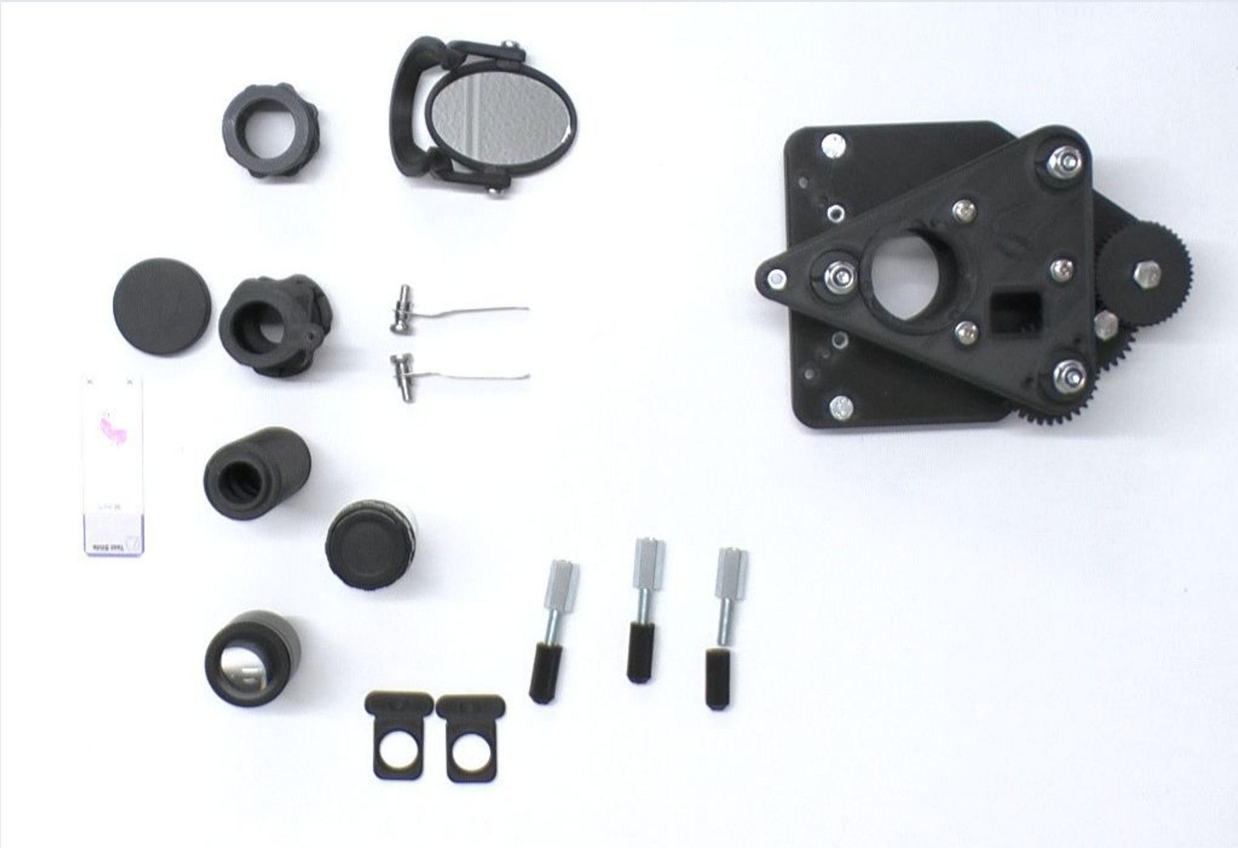
Assembling the Microscope

The following describes the recommended procedure for assembling your Foundation scope for the first time after removing all the parts from the shipping box. A video has been prepared to illustrate this and can be accessed via this link:

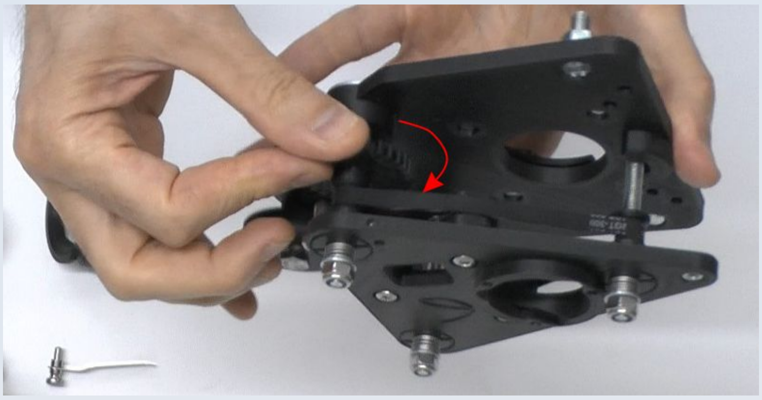
PUMA Assembly Video

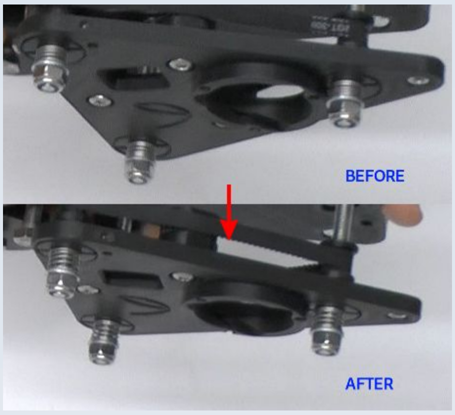
https://youtu.be/C-2vRsHi46c

Step 1. First remove any components from their separate bags / pouches and remove the ocular lens from its box (but keep the objective lens in its case for now)

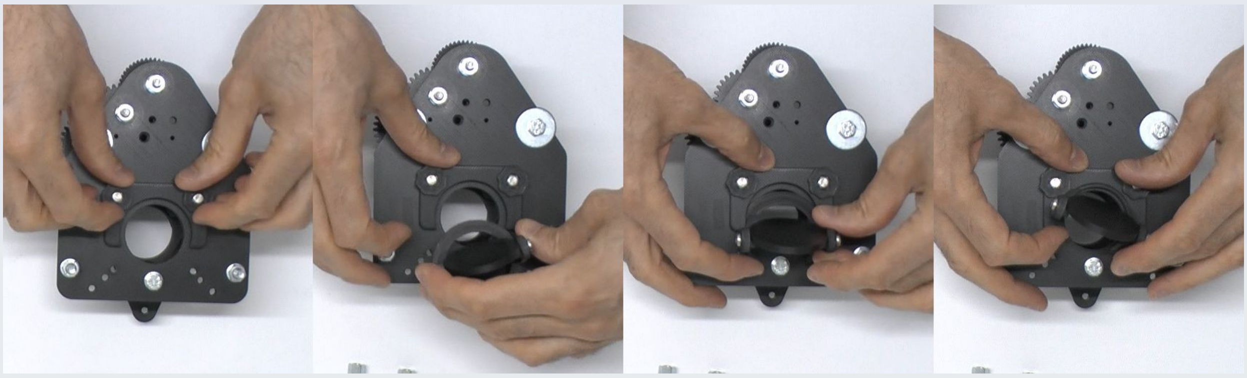


Step 2. When the scope ships from OptArc the focus platform is fully raised so as to accommodate the various parts that are packed in between the stage and the focus platform. This means the focus mechanism springs will be fully compressed so the next thing to do is lower the focus platform a little to take some of the compression off these springs. Do this by turning the coarse focus wheel a few turns as shown in the figure. When looking at the scope from the top aspect this amounts to clockwise rotations of the coarse gear. Don’t lower the platform all the way down. You will adjust correct focus after the scope is fully assembled (as will be described later).

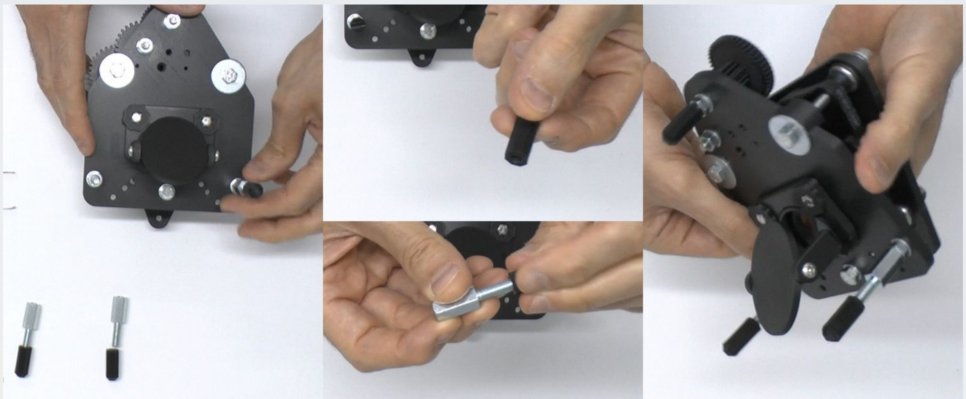




Step 3. Turn the scope upside down and loosen the thumbscrews that hold the mirror-to-baseplate attachment in place. Do not remove these thumbscrews, just loosen them enough so you can slide the mirror attachment into the slot between the baseplate (i.e. the undersurface of the stage) and the mirror-to-baseplate attachment – as shown. When inserted re-tighten the thumbscrews just enough to prevent passive wobble of the mirror attachment but loose enough to allow rotation of the mirror attachment.



Step 4. Screw on the front two legs which have the plastic foot part pre-threaded onto the metal spacer. Make these finger tight. For the back leg you choose whether you want to use the plastic foot only – this will allow the scope to tilt backwards for ease of manual viewing – or use a metal spacer + plastic foot combination (as you used for the front legs) – this will make all 3 legs of equal height so help keep the stage level (useful if you are using fluid specimens or oil immersion). To make the metal spacer + plastic foot combination first attach the plastic foot to the spacer (screw it onto the spacer all the way it can go) so that it will look identical to the front two legs.



Step 5. Insert the (empty) filters into the simple filter block. Note that the slots for these two filters have no dividing wall between them so take care to insert each filter level and not at an angle to cross over from one filter slot to the other. The filters must also be inserted the correct way up – the marking on the filter handle must face upwards (towards the ocular end of the optical tube) as shown in the figure. It is advised to insert the lower filter first because you can use the base aspect of the filter block as a guide to prevent angling the filter upwards across into the upper slot. The top filter can then be inserted and so the already sited lower filter will help prevent angling the top filter downwards:

Step 6. Screw the monocular tube into the top of the simple filter block by means of the smaller thread on the monocular tube and make this finger tight. **Do not over-tighten or the ocular tube thread may break**.

Step 7. Screw the ocular holder assembly onto the top of the monocular tube.

Step 8. Remove the objective lens from its protective case and thread it into the thread at the bottom of the simple filter block.

Step 9. Insert the ocular lens into the ocular lens holder – it is a simple push fit. This may not be a tight fit so **be careful not to invert the optical tube from now on or the ocular lens may fall out and be damaged**.

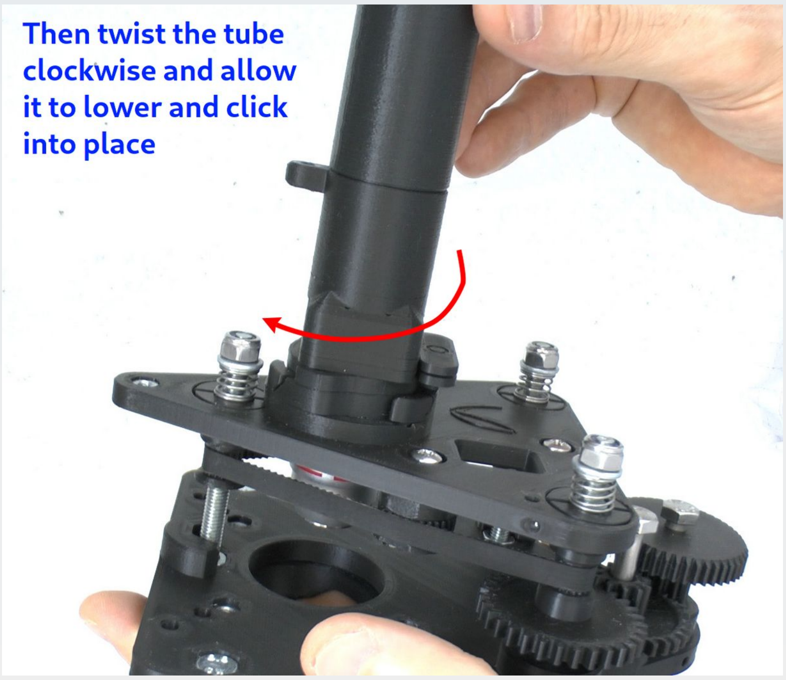
Step 10. (Refer to illustrations on next page). Now insert the completed optical tube into the focus plate of the stage via the quick release (QR) mechanism. The procedure described here was designed to minimise the risk of crossing the thread of this mechanism because forcing this in with a crossed thread could damage it.

First note that the final position of the optical tube, when fully inserted, is with the lug on the simple filter block facing forward (the lug it is slightly off centre by design so don't be concerned about that) and with the filters and filter slots facing back towards the focus gears (see picture). However this QR fitting is a rotational mechanism so start with the optical tube orientated 90 degrees anticlockwise so the lug on the filter bock faces left (as shown) and lower the optical tube into its receptacle hole in the focus platform till the tips of the three threads stop it going down any further. **At this point ensure that the optical tube is flat against the focus platform** (i.e. fully perpendicular to the focus platform, if the focus platform is tilted backwards then the optical tube must be equally tilted – it must be perpendicular to the focus platform, not the work bench). Now rotate the optical tube – while keeping it flat and level – clockwise. If all is well the optical tube tube should easily rotate and descend and click into place when a quarter turn is complete. If you find you are requiring force you should un-rotate and double-check to ensure the threads are not crossed (the video should make this clear if the below diagrams are insufficient).


FRONT
BACK


Begin insertion with the optical tube rotated about 90 degrees anti-clockwise so the lug faces to the left.


Ensure the tube rests flat against the focus platform

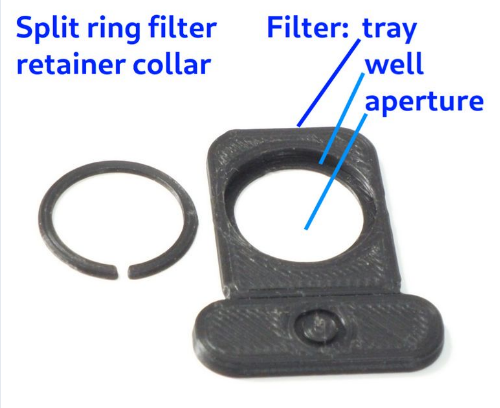

Then twist the tube clockwise and allow it to lower and click into place

Releasing the optical tube from the focus platform is simply a matter of rotating it anticlockwise.

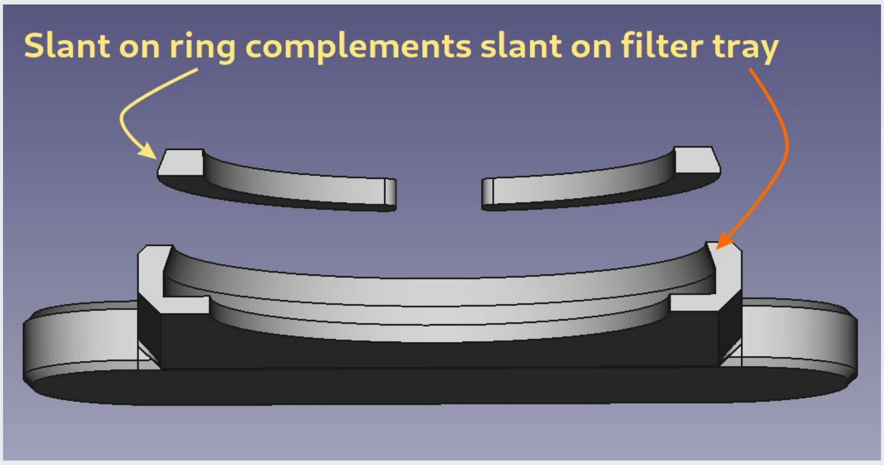
The only thing left to do now is add a specimen slide and adjust the focus. These steps will be elaborated in the next chapter because they are common to making observations.

Filter slider trays and retention collars

The filter trays supplied with the Foundation scope are empty (they don't contain any filter) but they are the complete mechanism whereby you may add your own filter. Each filter has a split ring filter retention collar. These can be kept separate or fitted to the empty filter trays as you prefer (see below on how to fit them). You will need to remove these retention collars if you plan to add an actual filter to the filter tray at some point.



To add a filter, your filter must be able to fit into the filter well in the tray without falling through the well aperture. This means it must be round with a diameter of 17 mm to 18 mm. Your filter must be no thicker than 0.9 mm or the total thickness of the filter tray mechanism once the retention collar is in place will be too thick to go into a filter slot in the filter block (you might get away with thicker filters if you only use the lower filter slot keeping the upper slot unoccupied but then you won't be able to properly retain the filter with the split ring filter retention collar).



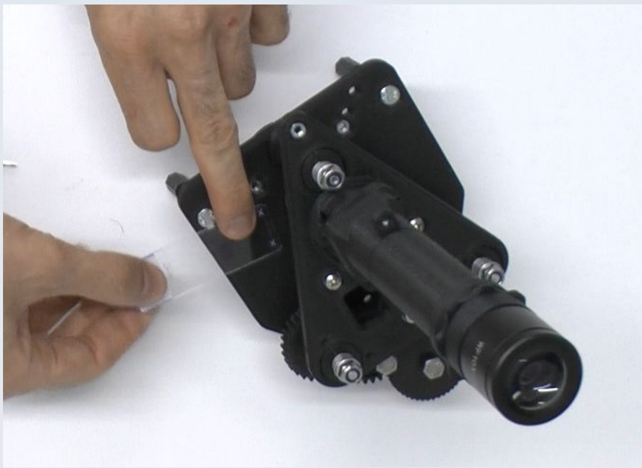
To fit the filter, ensure the collar is out of the filter tray, simply drop your filter in the filter well then re-apply the split spring collar to hod it in place. **Note that the collar MUST be fit the correct way up because it is not symmetrical**. The wall of the filter well and the wall of the split ring collar each have a complementary slanting angle to facilitate the collar's retention in the filter well (see picture of the cut-away CAD model above). To remove a filter retention collar you can use a needle or a tiny jeweller's flat bladed screwdriver to winkle it out of the filter well (taking care not to damage any filter that may be present). If there is no filter present you may be able to pull the collar out with your bare finger (no tools needed).

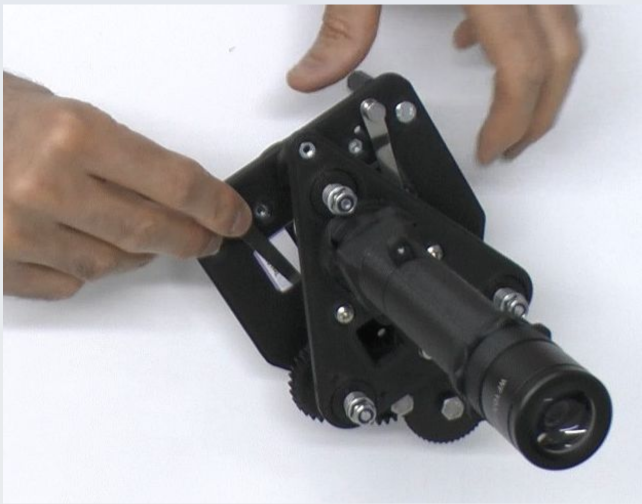
Making Observations

How to place a specimen on the stage

Although various types of specimen can be used this guide refers to the use of a standard 25 x 75 mm glass microscope slide containing a coverslipped specimen such as the test slide supplied with your PUMA Foundation scope.

Ensure the slide is the correct way up (label and coverslip facing up) and put it flat on the stage to one side of the objective then slide it on the stage till the specimen on the slide is approximately centred under the objective. This procedure is recommended to avoid the danger of scratching the objective with the slide – keeping the slide flat on the stage at all times reduces that possibility.



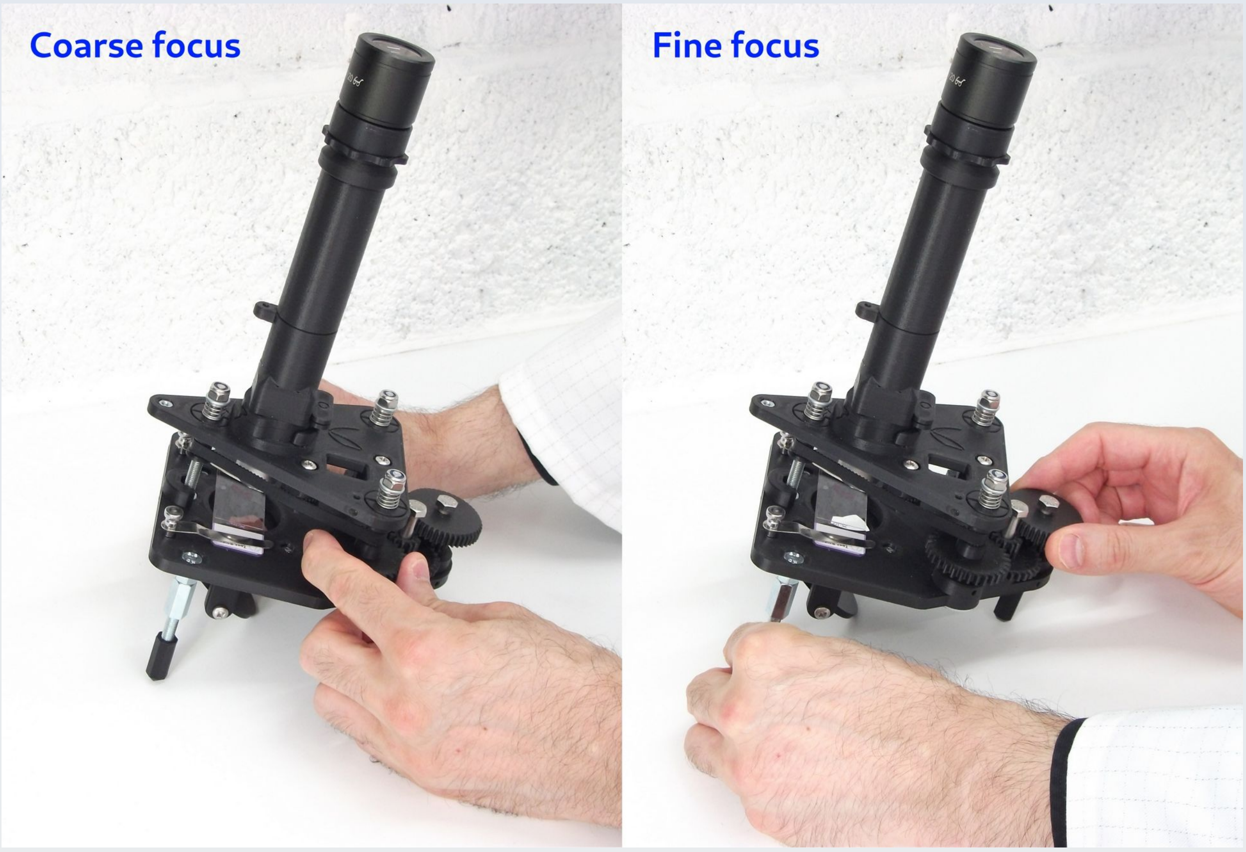


Once the slide is in position place a stage clip in its hole in the stage (these are the holes closest to the front edge of the stage) and press it down on the slide. **It is best to do it this way rather than fix the stage clip on away from the slide and rotate the clip onto the slide because that method will create tiny glass fragments as the metal clip scrapes over the edge of the glass slide and this glass dust is a health hazard and also could cause scratches on your optical lens surfaces over time**. The online video illustrates this safe method and how it differs from the method to be avoided.

Note that the specimen slide could alternatively have been inserted with the optical tube removed. This is sometimes preferred, especially when using high power (high magnification) objectives. High power objectives are physically longer than the x4 objective that is supplied with some Foundation scopes (as per purchase options) and so they have less clearance space between the objective lens and the stage. This means there is a higher risk of doing damage to the objective by touching it with the slide when inserting the slide with the optical tube in place.

How to focus the scope

You are now ready to focus the scope onto the specimen and begin observations. Position the scope so that the mirror can catch reflected diffuse light (e.g. from a cloudy sky) and, while looking down the ocular lens, hold on to one or more of the legs of the scope and rotate the focus gear (fine or coarse) till the image of the test specimen comes into focus. It is likely that, if you have followed this guide for assembling the microscope, the focus platform will be too high and so will need to be lowered. Therefore you can begin by rotating either focus gear in a clockwise direction. ***Due to the elastic backlash of the mechanism you may need to focus just past best focus and allow the backlash to take you back to best focus***.



Coarse focus may be quite hard to move in some cases. If that is the case you can perform all focus action with the fine focus gear – it will just take more turns to reach your destination.

Attaching a camera

A camera or camera adapter is not supplied as part of the standard PUMA Foundation scope package but with suitable additional equipment it is possible to take photos and video using PUMA. OptArc can supply a range of cameras suitable for use with any PUMA microscope. This section will give a brief overview of the use of three different types of camera. Detailed instructions will be left to the instruction manuals supplied with the camera or adapter equipment. Digital cameras require a computer and software to record the image but for some (e.g. smartphones) the computer and software are integrated into one device.

Eyepiece Camera

OptArc wide field eyepiece camera

Eyepiece cameras simply fit over the existing ocular lens and act as a recording surrogate for your eye. OptArc provides a 5 megapixel USB2 wide field camera which was designed specifically to be used with PUMA. Other manufacturers also provide eyepiece cameras however these may or may not be suitable for use with PUMA depending on the details of their lens angle, optical axis centration abilities, focus characteristics and whether they physically can fit over the PUMA ocular lens without damaging it. These are the issues you would need to research if you decide to use a non-OptArc eyepiece camera.

C-Mount Camera

C-Mount camera replacing eyepeice

C-Mount cameras expose the bare chip to the primary image of the objective, without an eyepiece in-between. This tends to give a small field of view with a highly magnified image. For this reason some form of beam size reduction optics are used. Either way, C-mount cameras need to have a C-mount to 23.2 mm barrel adapter to be used with a PUMA microscope and they fit into the ocular holder assembly in place of an ocular lens (i.e. the ocular lens must be removed).

Smartphone Camera

Eyepiece smartphone adapter

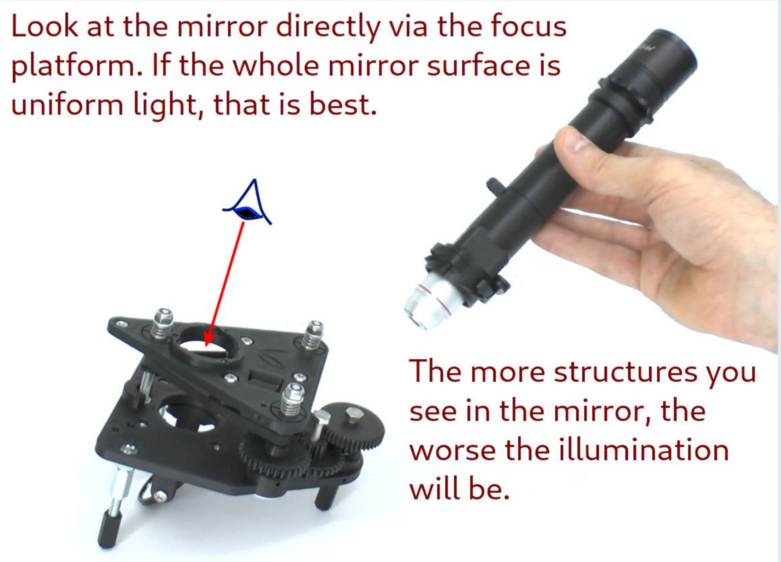
A smartphone camera can be used by means of a smartphone-to-eyepiece adapter (as shown in the figure). These adapters fit over a standard ocular lens and allow the smartphone to be held in such a way as to centre its camera lens over the microscope's ocular lens. The smartphone camera system is then used as normal to take the photos. It is advised that, due to the fact that pressing the shutter button on a camera attached to the scope will cause some vibrations in the image which need time to settle down, a time delay shutter setting should be used with a delay of at least 2 seconds. Because these adapters hold the smartphone off to one side this could imbalance the optical tube and the scope as a whole. For this reason it is advisable to use the full hind leg of the scope (including the metal spacer) and also use some form of additional custom strut support to prevent the weight of the camera bending the optical tube off-axis.

Advice for Good Illumination

For optimal illumination aim to fill the mirror surface with even bright light but not too bright so as to damage eyesight. **Avoid using direct light from the sun or sunlight reflected off a shiny surface. Avoid using lasers as a light source if you are going to look down the scope by eye** (as opposed to only using a camera for observation).

The aim mentioned above is important because, especially with low power objectives such as a x4 objective, any structure to the light (such as images of window frames or curtains or some outdoor scenery) can, albeit blurringly, superimpose itself on your observed image. Likewise you should avoid using strip lights or spotlights because these will not uniformly fill the mirror.

A good way to tell if your illumination is diffuse and extensive enough is to remove the optical tube and look directly down at the mirror through the main central hole in the focus platform. If you see structures other than diffuse light all across the mirror then you do not have ideal illumination. You can get away with some laxity in this rule but the more structure you see in the light the worse the final image quality will be.



An ideal source is a cloudy daytime sky with the microscope outdoors or very close to a large window (the further you are from the window the more of the surrounding structure will be captured by the mirror such as window frame, etc. and we have already noted that structured illumination is suboptimal).

The PUMA Foundation scope mirror is articulated so it can be conveniently directed to a light source even if the scope itself is restricted from being pointed in that direction for any reason. However when directing the mirror to a light source try to avoid getting any of the legs of the microscope in the path of the light between the light source and the mirror because the shadow / reflection of the leg will degrade the quality of the illumination.

If using the scope with an artificial light source try to get the illumination as diffuse as possible and to cover as much as possible of the surface of the mirror. This will require either a very large illuminated surface or a smaller diffuse illuminated surface which is very close to the mirror. In some cases it may help to cover the entire surface of the mirror with a uniform diffuser sheet such as opalescent plastic but if you use anything with a texture to it then that texture may be seen superimposed on the image of your specimen.

If using higher power objectives, especially more than x10, you will notice the image to be ‘tinny’ (lacking in good plain area contrast but having excessive edge-type contrast) and / or dark. This is because you require diffuse light to enter at a larger range of angles for higher power objectives and the best way to achieve this is with a condenser module such as the optional PUMA Abbe condenser upgrade.

Another advantage of using the PUMA Abbe condenser upgrade module is that is comes with a filter slot which allows you to shape the Fourier aperture of the condenser so you can achieve advanced imaging modalities such as dark ground (dark field) microscopy and Schlieren phase contrast. These can be achieved with simple fixed filters and do **not** require the use of the electronic spatial light modulator (which is a more advanced optional extra). The next chapter gives more detail about optional upgrade modules.

Upgrades

The PUMA Foundation scope is the simplest functioning microscope that can be made with PUMA modules. It requires no electronics and is very light and portable. However, the functions of the Foundation scope can be greatly extended by the addition of any one or more of many optional upgrades thanks to the modular nature of PUMA. At OptArc we aim to make some of the more popular upgrade modules available ready made but **we cannot guarantee that any of the upgrades discussed below will be commercially available from us** – in that case you may have to build them DIY using the PUMA open source specifications. These upgrade modules are briefly discussed here but more detail can be found on the OptArc website (for ready-made upgrade kits) and in the Journal of Microscopy publication, the PUMA GitHub page and the PUMA Microscope official YouTube channel. Links to these sources are given below:

PUMA Overview Video	PUMA GitHub Page	PUMA Journal Article
https://youtu.be/7UbkrZyNgpo	https://github.com/TadPath/PUMA	https://pubmed.ncbi.nlm.nih.gov/34151425/

Higher Magnification

Total magnification is the magnification of the objective multiplied by the magnification of the ocular. For example a x4 objective and x10 ocular gives 4x10 = 40 times total magnification.

PUMA uses standard RMS-thread objectives which have a mechanical tube length of 160 mm and a conjugate focal distance of 195 mm. You cannot use 185 mm conjugate focus objectives with PUMA. You can use 195 objectives with a 170 mm mechanical tube length (such as old Leitz objectives). These can be used as is with the standard OptArc PUMA Foundation scope but they will not be par focal with 160 mm tube length objectives. However, to properly use 170 mm objectives, and especially if you want to use them with more advanced options such as the binocular head or AR HUD projection system, they you should use the optional longer ocular cap specifically designed to accommodate 170 mm objectives. This 170 mm ocular cap is an optional upgrade module, not supplied as standard with the OptArc Foundation scope package. If you want to use infinity optics lenses, this is possible but only with the addition of an optional 100 mm focal length tube lens fitted inside the optional advanced filter block module. This infinity optics upgrade is a concession – to allow you to use any infinity objective you happen to have lying around – and is not a recommended upgrade because 100 mm focal length is too short for optimal image quality with most infinity objectives. It ‘works’ but you get exaggerated lack of planarity of the image. This is because most infinity optics tube lenses have a much longer focal length of about 250 mm so the short 100 mm tube lens of the current PUMA infinity module does not allow enough distance for the planarity and colour correction paths of the infinity objectives to fully work. I am currently working on designs for a more dedicated high quality infinity optics adapter for PUMA but this is still at the design stage at the time of writing this manual and not currently available as a upgrade.

You can use objectives of up to x100 oil immersion with a PUMA system but in order to get a bright clear image with any objective higher than x10 you are advised to also use the Abbe condenser upgrade discussed below.

You can also use higher magnification oculars such a x20. Thus the total magnification achievable is between x1000 and x2000 using a x100 oil immersion objective and either a x10 (the standard) or x20 (an upgrade) ocular.

Be advised that if you use a higher magnification ocular you will see a smaller field of view. In particular you will not be able to see the whole augmented reality heads up display (AR HUD) interface if you use a x20 ocular.

OptArc supplies high quality Plan Achromat objectives but you can also use legacy objectives from Olympus, Zeiss and Leitz as well as other professional microscope manufacturers.

Beware of cheap generic lenses. Many other brands or generic RMS objectives can also be purchased very cheaply however those cheap objectives tend give lower quality images, in particular the field of view will not be flat (uniformly in focus) from the centre to the periphery. The cheap lenses will show either the central area in focus and the periphery blurred or vice versa depending on the position of current focus. Cheap oculars can also be purchased but these too suffer from a variety of quality issues and often have a small exit pupil with short eye relief meaning you will need to get your eye very close to the tube to see the whole field of view which can be uncomfortable for long term viewing and impossible for people who wear glasses unless they remove their glasses (then the focussing will need to be adjusted and this may cause other problems such as worse field curvature and lack of colour registration). PUMA was designed to be a professional microscope but if you use cheap toy lenses with a PUMA system then your viewing experience will be like that of a cheap toy microscope.

Long Legs System

The long legs upgrade allows greater clearance under the microscope and is required for illumination system upgrades (including the addition of an Abbe condenser) with the exception of epi-illuminators.

Abbe Condenser

The PUMA Abbe condenser module allows high magnification, high numerical aperture (NA) imaging by gathering the incoming light and dispensing it in a wide angled beam through the specimen very close to the objective lens input aperture. It can also be oiled so as to provide double-surface oil immersion for the highest NA lenses (this means that oil can be put on the condenser lens to optically connect the condenser to the undersurface of the glass specimen slide as well as putting oil on the coverslip of the slide to optically connect it to the objective lens).

The NA of the condenser itself is 0.938. While this is lower than the NA of most oil immersion lenses (typical NA = 1.25) it nevertheless gives a good quality image in practice when used with such lenses.

The PUMA Abbe condenser can be used with either the mirror illuminator of the original Foundation scope or the Köhler illuminator upgrade module which uses a powered LED for illumination.

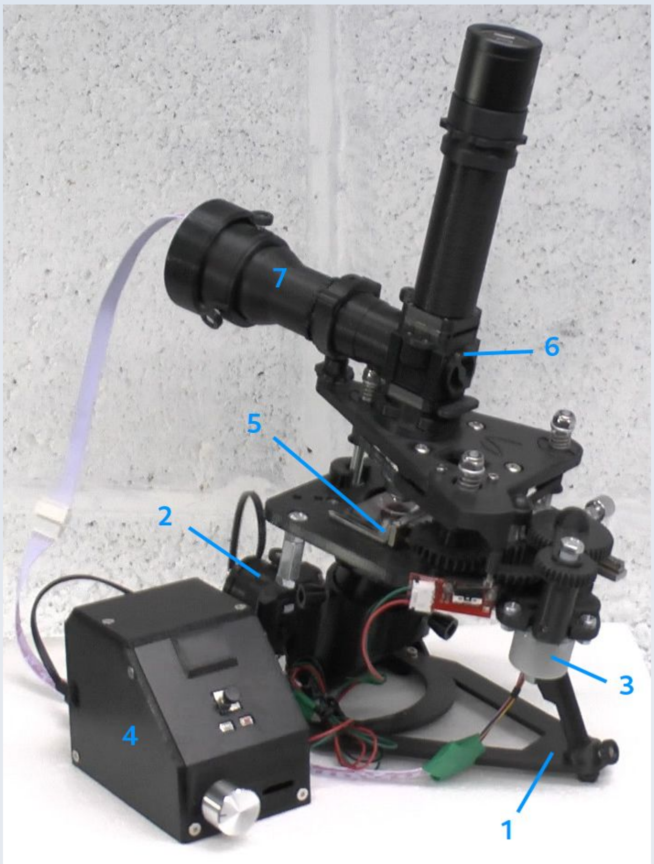
Regardless of which light source is used, the PUMA Abbe condenser module has a filter slot which allows a filter-type aperture to be used to shape the aperture of the Fourier plane of the condenser and so provide for advanced illumination effects such as dark ground microscopy (= dark field microscopy), Schlieren-type phase contrast (using normal objectives, not phase objectives), Rheinberg filters and more. These effects can be achieved with fixed 3D-printed plastic filters.

Those imaging modes can also be achieved with the additional optional upgrade modules of the PUMA TFT-screen based spatial light modulator (SLM) and PUMA Control Console but those advanced devices are not necessary for static effects, being more appropriate if active computer control of the condenser aperture is required for more advanced projects.

One limitation of the PUMA Abbe condenser is that the lenses display some polarisation birefringence meaning that you cannot use the condenser for trans-polarisation microscopy. For polarisation microscopy a dedicated illuminator module is available (see below for details).

Köhler Illuminator

The PUMA Köhler illuminator kit allows upgrading the microscope to use a powered LED illuminator thereby freeing you from being dependent on finding a good external light source (as you are with the original Foundation scope mirror system). It also gives more control over the intensity of the light source and its wavelength because the LED used can be obtained in a variety of wavelengths as optional extras.



Multiple upgrades applied as follows:

1. Long legs system
2. Köhler illuminator
3. Motorised Z-stage
4. PUMA Control console
5. XY slide holder
6. Advanced filter block
7. AR HUD system

The light source LED can be powered using the simple PUMA Lite battery-operated control box or the larger (but also portable) PUMA Control Console which, in addition to powering the LED like the PUMA Lite, also sports an Arduino Nano microcontroller and multimedia interface to allow TFT screen-based devices and the (optional) stage Z-motor to be controlled. Both the PUMA Lite and the PUMA Control Console can also be powered by an external mains power supply if full portability is not required.

The Köhler illuminator offers access to the illuminated field stop which is also controlled by filters that are cross-compatible with the filters used for the condenser aperture described above (the illuminating aperture stop).

The standard Köhler illuminator gives good uniform illumination for objectives of x10 to x100 oil. The field of view of most x4 objectives is a little too wide for the standard Köhler setup and will show some darkening at the extreme periphery of the field but is still useable. If you really need full Köhler flat field illumination with a x4 objective lens then there is an optional upgrade to the standard Köhler illuminator which will allow that.

One limitation of the PUMA Köhler illuminaton / condenser system is that the lenses display some polarisation birefringence meaning that you cannot use it for trans-polarisation microscopy 'as is'. For polarisation microscopy a dedicated illuminator module is available (see below for details).

Trans-Polarising Module

A special powered LED illuminator module is available to allow trans-polarisation. This works as an alternative to the PUMA Abbe condenser and PUMA Köhler illuminator because those two modules can't be used for polarisation.

The PUMA Trans-Polarising illuminator can be used with objectives up to x40. Higher magnification objectives may be used but the image may not be optimal in resolution and contrast.

Advanced Filter block

The PUMA advanced filter block is a direct replacement to the standard filter block. It has a 50:50 beam splitter in its body, an upper filter slot above the beamsplitter (in addition to the 2 lower filter slots identical to those on the standard filter block) and a receptacle for an optional tube lens to permit infinity objectives to be used.

The advanced filter block allows the use of the PUMA augmented reality projector module for heads-up display. It also allows the use of the epi-illumination and trinocular port options described below.

The infinity tube lens option is a concession to design and is not ideal because it has too short a focal length (100 mm) compared to the focal length of most infinity microscope tube lenses (about 250 mm). This means that infinity objectives used with this option show a high degree of field curvature. A more optimised infinity adapter is currently in development. However, the cavity provided for this infinity tube lens may alternatively be used for any custom optical element that can fit in the cavity.

Epifluorescence upgrade using the advanced filter block and including the PUMA Lite controller.

Epi-illumination

Three powered epi-illumination modules are available – all require the use of the advanced filter block. These are:

1. Epi-illumination alone
2. Epi-polarisation
3. Epi-fluorescence

All use the same LED lamp holder as the Köhler illuminator and the trans-polarising illuminator so LEDs of a variety of wavelengths can be used.

XY Mechanical Vernier Slide Holder

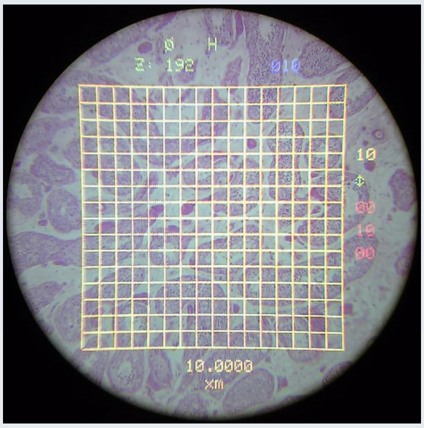
The stage clips can be replaced by a mechanical slide holder with rotary controls to move the slide in X and Y and with a Vernier scale on X and Y for ease of slide screening and manual measurements of specimen feature dimensions.

Trinocular Port

The PUMA trinocular port module requires the use of the advanced filter block and adds an adjustable ocular viewport to the back of the filter block. This ocular can be used for direct vision but is more likely used to affix a camera for recording or broadcasting observations which a user can observe simultaneously via the main ocular head.

Furthermore this viewport is designed to attach via a rotatable polariser so any image overlay from a simultaneously attached AR HUD module can be optically erased if desired (as well as having the option of using this port for trans-polarisation viewing).

Augmented Reality (AR) Projector for Heads-up display (HUD)



The PUMA AR HUD module superimposes the image of a digital screen onto the live optical image and allows advanced features such as an interactive pointer, image analysis, quantitative microscopy, timing live cell events, and more. This is controlled by the PUMA Control Console and requires the use of the advanced filter block.

Spatial Light Modulator (SLM)

The PUMA SLM is a semitransparent TFT screen filter that is computer controlled (such as with the PUMA Control Console). It acts as an electronic active matrix filter and is most powerful when used in the Fourier plane of the Abbe condenser providing adaptive apertures for modalities like Schlieren phase contrast, dark ground microscopy, Fourier ptychography and 3D tomography amongst other things.

Note that the PUMA Control Console can only control one TFT-based device at a time so if you are using both the AR HUD and SLM simultaneously you will need two controllers.

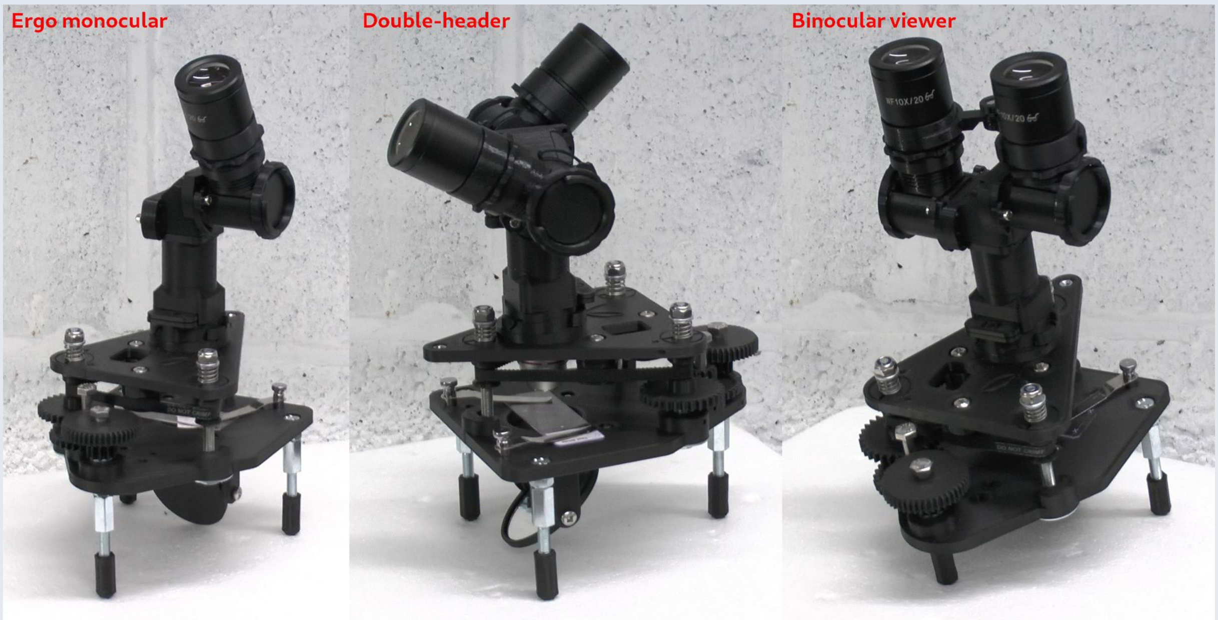
Binocular Head

The PUMA binocular head module can work with either filter block (standard or advanced) and can be configured to provide any one of three modes of viewing as shown in the figure on the following page):

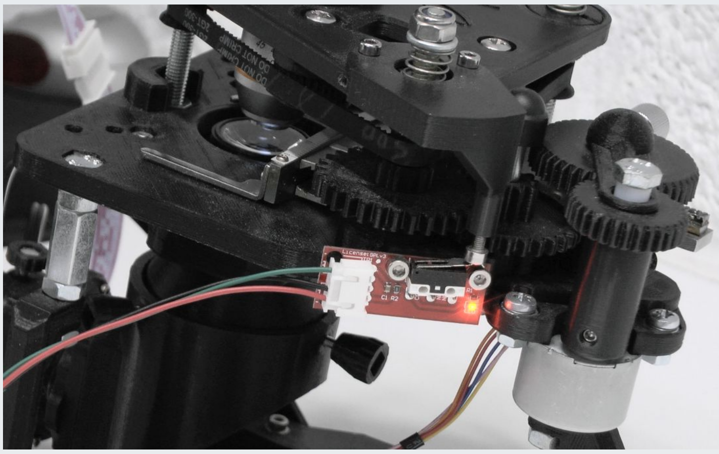
1. Monocular ergo-head (i.e. a monocular viewport with an adjustable angle – useful for comfortable ergonomical viewing if you have to have the scope level instead of inclined backwards). This mode only uses one of the two oculars for viewing, the other can be capped off (so there is no second ocular at all) or may be used, e.g., to fit a camera to.

2. Double-header viewing. In this mode each of the two oculars is tilted in opposite directions so two people can share the same microscope for discussion of the observations (which can be facilitated by use of the AR HUD pointer).

3. Single viewer binocular vision. This allows a user to use both eyes to make observations which is more comfortable especially for prolonged observations.



Z-Motor



The focussing mechanism can be motorised by use of the PUMA Stage Motorisation module. Use of a motor to focus the stage is particularly helpful in reducing the image ‘wobble’ that is noticeable when doing manual focussing with high power objectives.

The stage Z motor is a stepper motor and must be powered by a suitable stepper motor driver logic circuit – it cannot be driven by simple electrical connection like a continuous DC motor. A suitable stepper driver is the ULN2003 Darlington pair IC. If you do not want to build your own stepper driver system then the PUMA Control Console can be used because this contains a ULN2003 stepper driver module as standard, as well as a friendly user interface with some advanced motor control functions built in to its Arduino Nano microcontroller.

Maintenance

This chapter discusses routine user maintenance. For details about how to fully take-apart your scope and re-assemble it see the PUMA GitHub page and PUMA Microscope YouTube channel videos.

Avoid excessive heat

The body of your microscope is made of PLA plastic which has a glass transition temperature of about 60 ºC (140 ºF). This means it will start to soften and may deform if allowed to get near that temperature. It is advisable to avoid exposing the microscope to temperatures in excess of 40 ºC (104 ºF).

Under no circumstances should the scope be placed on or near a room heater or radiator (e.g. to dry off if it gets wet or for any other reason).

Avoid keeping the scope in a vehicle on a sunny day or in a transparent closed cabinet exposed to the sun.

Moisture and condensation

The body of your microscope is made of PLA plastic which can withstand moisture by water and many solvents but it is not completely solid or waterproof meaning that if submerged to exposed to excessive wetness that liquid might find its way into the air cells inside microscope components and take a long time to evaporate. Furthermore PLA plastic is biodegradable so having aqueous solutions stagnating inside its components could hasten degradation (although noticeable effects may take several years).

For these reasons please protect your scope from the rain and do not immerse it in liquid.

The lenses of the microscope should also be protected from moisture and condensation because if any water or other solution gets in-between. lens elements the optics may become unusable because trapped moisture can cause condensation on the inner, unexposed, layers of the lenses and this cannot be wiped off. It may take considerable time to evaporate. and in that time fungal attack on the sensitive anti-reflection coatings may build up and permanently damage your lenses.

Lens care

The need to keep lenses away from moisture and condensation was stated in the preceding section – which see for details.

Take care to avoid physical contact with lens surface as these have sensitive anti-reflection optical coatings that may be damaged with coarse contact and lenses may get chipped which will reduce their image forming quality.

Avoid hard knocks or dropping the lenses (especially the objective lenses) as this can dislodge elements from precisely aligned positions and make focal planes uneven (images will not appear in focus throughout the image plane regardless of how you adjust the scope)

When not using the microscope for a short period of time, place the passive dust cap over the ocular lens to prevent dust build up.

If not using your microscope for longer periods consider using a plastic dust cover to cover the whole scope and so prevent dust build up on the stage and focus gears as well (alternatively keep it in a cabinet but avoid transparent cabinets exposed to sunlight or there is a risk of heat deforming the microscope – see section on 'Avoid excessive heat' above).

If not using the scope for more extended periods of time it should be field-stripped and packed away in a soft case (see the You Tube video on how to field strip a PUMA microscope) and kept in a cool dark place away from moisture / condensation.

To clean the lenses or other optical surfaces use an air duster or pneumatic soft lens brush to blow away any surface debris first. Then use only a special purpose lens cloth with lens cleaning solution. Wipe gently and carefully to avoid scratching the lens coatings. It is best to avoid any wiping with first surface mirror elements such as in the advanced filter block or binocular head module unless absolutely necessary (an air duster should be sufficient – tiny particles remaining should not cause significant image degradations and the risk to damaging the mirror coatings outweighs the benefit of removing such particles).

When using oil-immersion objectives only use synthetic immersion oil that is designed for use with microscope objectives (such as OptArc immersion oil) because other types of oil may damage the lens surface or cements used in fixing the lenses in place. Do not let oil seep into the spring mechanism of the objective (so keep an oiled objective facing downwards until you have wiped the oil off it). Wipe oil off an oiled objective immediately after observations have ceased using a special purpose lens cloth / lens paper prior to storing the objective in its protective case. Do not use any kind of oil or other immersion medium with lenses that are not specifically designed for immersion use.

Keep all unused objectives in their protective cases when not in use. If placing an objective lens on a work surface. temporarily ensure that surface is free of dust and place the objective base-side down (i.e. with the threaded end of the objective on the surface). Do not leave objectives exposed on a surface like this for more than a few seconds (it may be necessary to do this while changing objectives).

Avoid letting the sun shine directly down the ocular lens of the microscope or on the microscope mirror when not in use.

Lubricating the focus mechanism

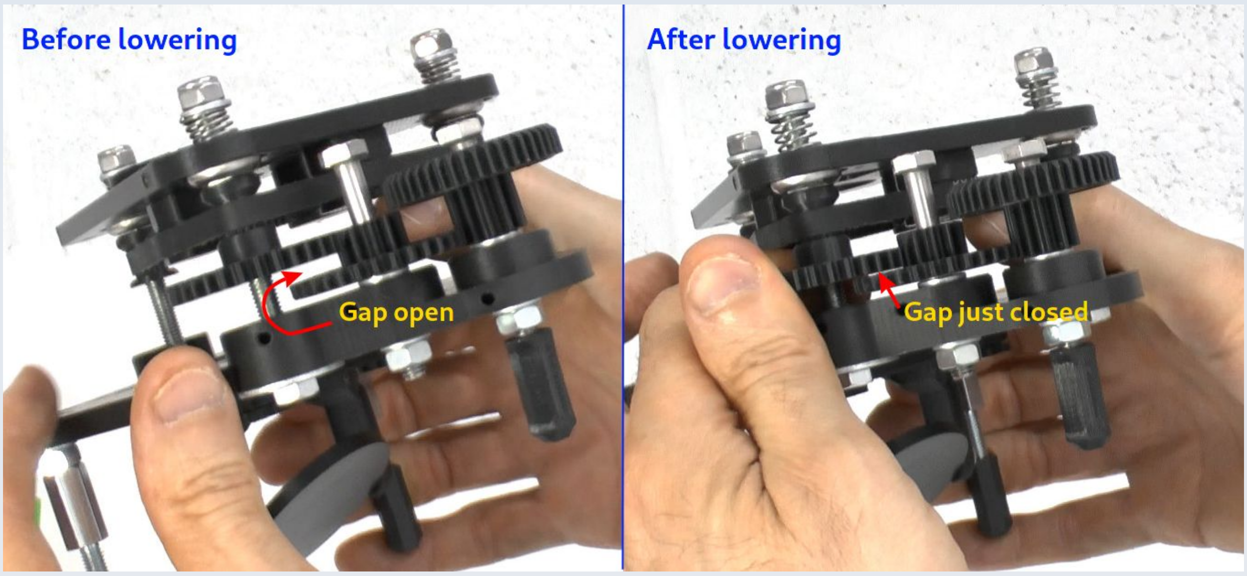
The focus mechanism should be greased occasionally to maintain smooth and predictable focussing behaviour. How often depends on how much it is used. Do this about once every 6 months if there is only light use and more often if the scope is used more extensively. If you notice the focus mechanism behaving erratically, getting stuck, etc. then this may be because it needs re-greasing.

For this purpose use a thick gel-like axle grease or petroleum jelly, etc. Runny oils (such as sewing machine oil) and WD 40 (or similar) are NOT recommended.

To apply grease:

Step 1. Remove the optical tube and slide and keep them safely to one side.

Step 2. Lower the focus platform as far as it will ***safely*** go. This will be when the large part of the coarse focus gear just touches the large part of the intermediate focus gear (see figure). **Do not try to lower the stage more than this or you will damage the gear system**.

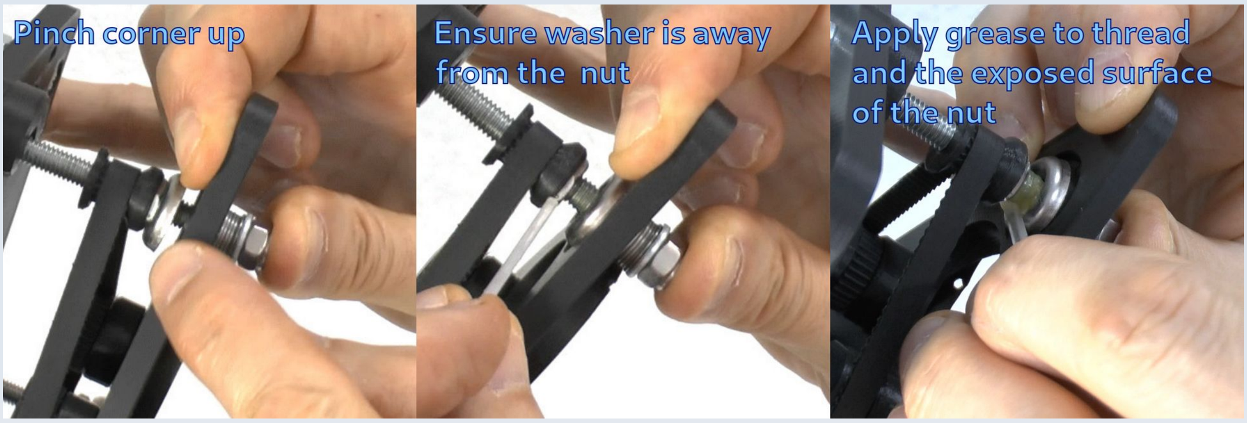


Step 3. For each of the the three corners in turn do the following (see picture on next page):

Step 3a. Hold the stage upside down and with the your thumb on the focus nyloc nut and your finger on the undersurface of the focus platform corner, squeeze your finger towards your thumb so that you elevate the corner of the focus platform by compressing the focus spring.

Step 3b. Ensure that the loose countersunk screw cup washer falls ‘down’ onto the undersurface of the focus platform so as to expose the surface of the nut in the focus pulley and the thread of the focus post bolt. With your free hand apply grease to the focus bolt thread and the surface of the focus pulley nut. It may help to use an applicator like a thin strip of plastic but whatever you use ensure that it does not have free surface debris that could get into the bolt thread (for this reason cotton buds should be avoided because loose cotton strands could build up in the mechanism).

Step 3c. Release the pressure you were applying to the focus spring so that the corner of the focus platform returns to its normal position and ensure that the countersunk screw cup washer rests evenly on the focus pulley nut (if it does not, re-elevate the corner by applying pressure to the spring as before and then release it again – repeat a few times until the washer lies evenly and flat on its focus pulley nut. You may need to give the washer a little nudge while doing this if it doesn’t settle by itself).



Step 4. When the above procedure is applied to all three corners you should elevate the focus platform by turning the coarse focus gear anticlockwise (as seen from above). Elevate it until the springs are almost fully compressed then lower it a few turns.

Your focus mechanism is now properly greased.

Adjusting the belt tension

The three eccentric belt tensioning devices are set to their minimum setting when the scope is built and this is perfectly adequate because at that setting a new timing belt is at its optimum tension level.

Too tight a tension and the stage will become difficult to move and XY hysteresis will be exaggerated. Too loose a belt can cause exaggerated backlash as well as XY hysteresis. So there is an optimum mid-range level of tension and this is the level of tension of a new belt with the eccentric belt tensioning devices set to their minimum setting.

Over a period of several years it may become necessary to tighten the belt a little but do not over-tension them. When adjusting belt tension note that each of the three arms of the belt should be of equal tension.

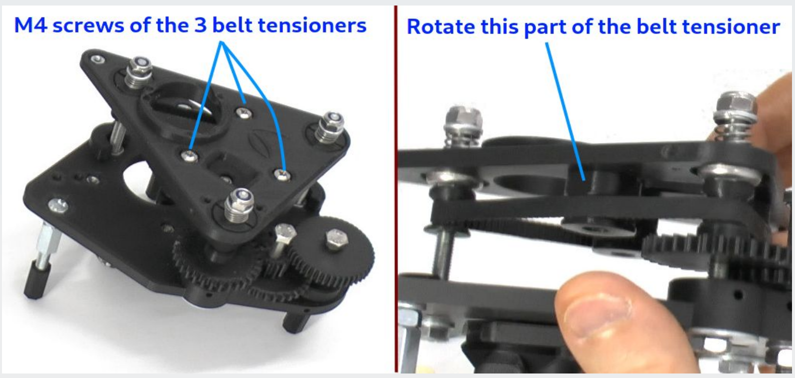
The procedure, performed for each of the three belt tensioners, is as follows:

Step 1. Remove the optical tube and slide and keep them safely to one side.

Step 2. Slightly loosen the M4 screw that holds the tensioner in place but do not remove the screw.

Step 3. With finger and thumb rotate the upper part of the belt tensioner device till you achieve the desired degree of tension. If it is too difficult to rotate the device loosen the screw a bit more and try again.

Step 4. When the desired tension position is reached, re-tighten the M4 screw.



Adjusting the optical axis of the optical tube

If the slide is perfectly flat on the stage and your specimen is also a perfectly flat specimen and the objective is properly threaded onto the optical tube but you see one side of the field of view in focus while the other side is not then the focus platform may nor be perfectly parallel to the stage/baseplate but you can compensate for that with the adjustable articulations.

However, altering the adjustable articulation should not normally be necessary. More likely the problem is caused by some other remediable issue. For example:

1. The actual specimen may not be perfectly flat.

2. There may be some grit or other particle under the slide (either stuck to the slide or stuck to the stage).

3. The slide is not being held perfectly flat against the stage. This is a particular issue with slides that are held with the mechanical XY Vernier slide holder upgrade (rather than the stage clips that come as standard with the PUMA Foundation scope) because the XY holder holds the slide slightly off the surface of the stage and its mechanism is easily tilted during normal use.

4. The optical tube is not fully rotated into the QR mechanism. that holds it to the baseplate. This can happen during normal use as one handles the optical tube.

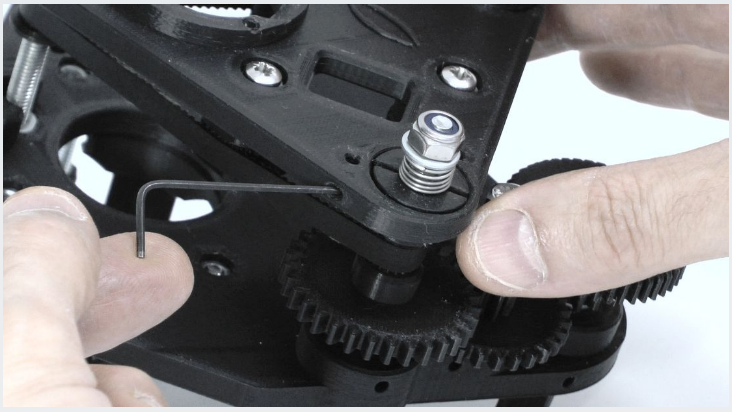
5. If you are observing with a camera it could just be that the camera is not fixed flat against the eyepiece or if the camera is heavy and unbalanced (e.g. if using a smartphone with an eyepiece adapter) it could be causing the eyepiece or optical tube to bend to one side therefore throwing the optical tube off axis.

For this reason if you feel the need to adjust the optical axis with the adjustable articulations, first check that none of the above causes are in effect and if they are then remedy them. It would be better to address the problem at source rather than alter the angle of the focus platform via the adjustable articulations.

If, after all the above has been checked and addressed, you still want to adjust the optical axis then the procedure is as follows.

Step 1. You can leave the slide on the stage and the optical tube in place. Looking at the slide in focus can help guide your adjustment as you make it.

Step 2. For the corner you want to adjust, first unscrew the fixing grub screw that holds the adjustable articulation in position. Do not remove the grub screw from its hole – just unscrew it a few whole turns.



Step 3. Using a special lens removal tool (or a partially open pair of scissors or stiff forceps) adjust the height of the articulation by screwing it one way or the other.

Step 4. When the desired level is reached re-tighten the grub screw. **Take care not to screw this in too much or it can damage the system and prevent it from being adjusted again**. You just want enough pressure to make it difficult to turn the adjustable articulation.

Disposal and Recycling

The optics are made of glass and metal (the metal for most optics being aluminium or an alloy thereof).

The metal fixings are made of steel (mostly stainless steel but some elements may be galvanised steel).

The 3D printed plastic parts of your microscope are made with poly-lactic acid (PLA) plastic. The following advice is current as of 2021. Consult your local authorities for the latest situation.

Recycling of PLA Plastic in the UK

PLA is compostable but most councils in the UK do not accept it in green or food waste.

PLA can be recycled but only by a very few specialist facilities. Most councils in the UK will not accept PLA plastic in their recycle bin waste.

PLA should therefore be disposed of in general household waste or sent to a specific facility that will accept it for recycling. Those with the ability to do so may also grind used PLA and reform it into usable 3D printer filament.

Recycling of PLA Plastic outside the UK

Please consult with your local authorities for recycling advice.

Electronics

The PUMA foundation scope has no electronic components.

Electronic components used in various upgrade modules should be disposed of in special electrical goods recycle facilities and household battery recycle facilities (for batteries).

Electronic components should NOT be disposed of in the general household waste or general household recycle bins.

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