INTEGRA



EVOLVE Manual Pipettes

Operating Instructions

INTEGR ∧ Declaration of Conformity

INTEGRA Biosciences Corp. - Hudson, NH, USA

declares on its own responsibility that the devices

Description	Models
EVOLVE Manual Pipettes	3011, 3012, 3013, 3015, 3016, 3018, 3019, 3022, 3024, 3025, 3026, 3028, 3032, 3034, 3035, 3036, 3038, 3042, 3044, 3045

comply with:

EU Regulations	Scope	Date effective
1907/2006	Registration, evaluation, authorisation and restriction of chemicals (REACH)	01.06.2007
EU Standards	Scope	
EN 9001:2015	Quality Management	

USA Regulations	Scope
17 CFR Parts 240 & 249	Dodd frank "Conflict minerals"
27 CCR Parts 25102- 27001	Proposition 65: The safe drinking water and toxic enforcement act
TSCA 40 CFR Part 751	Toxic substances control act

International Standard	s	
ISO 8655-2	Piston pipettes	

Zizers, 2023-12-08

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1 Introduction

These operating instructions contain all the information required for installation, operation and maintenance of the EVOLVE manual pipettes. This chapter informs about the symbols used in these operating instructions, the intended use of the EVOLVE manual pipettes and the general safety instructions.

1.1 Symbols used

The operating instructions specifically advise of residual risks with the following symbols:



WARNING

This safety symbol warns against hazards that could result in injury. It also indicates hazards for machinery, materials and the environment. It is essential that you follow the corresponding precautions.



CAUTION

This symbol cautions against potential material damage. Follow the instructions.



NOTE

This symbol identifies important notes regarding the correct operation of the device and labor-saving features.

1.2 Intended use

This is a general-purpose laboratory instrument for use in research only. Any use of this instrument in a medical or IVD setting is under the sole responsibility of the user.

EVOLVE manual pipettes are intended to be used in a laboratory for aspirating and dispensing liquids in the volume range of 0.2–5000 μl using GRIPTIP pipette tips.

1.3 Safety notes

EVOLVE manual pipettes comply to the recognized safety regulations and is safe to operate. EVOLVE manual pipettes can only be operated when in perfect condition and while observing these operating instructions.

The device may be associated with residual risks if it is used or operated improperly by untrained personnel. Any person operating the EVOLVE manual pipettes must have read and understood these operating instructions, and particularly, the safety notes, or must have been instructed by supervisors so that safe operation of the device is guaranteed.



WARNING

When handling dangerous substances, comply with the material safety data sheet (MSDS) and with all safety guidelines such as the use of protective clothing and safety goggles.

Only use GRIPTIPS® brand pipette tips to ensure the proper function of the EVOLVE manual pipettes and to comply with the general warranty conditions. Damage to the pipettes and risk to operator's health and safety may result from using non-specified pipette tips.



CAUTION

- Do not immerse the EVOLVE manual pipettes in liquid. The fluid can damage internal parts. Avoid pipetting of liquids whose vapors could attack the materials PA (polyamide), POM (polyoxymethylene), FPM (fluor-rubber), NBR (nitrile-rubber), CR (chloroprene), silicone. Corrosive vapors could also damage metallic parts inside the device.
- Repairs may only be performed by INTEGRA Biosciences AG or by an authorized after-sales service member.
- Parts may be replaced with original INTEGRA Biosciences parts only.



NOTE

Prolonged exposure of the EVOLVE manual pipettes to UV-light can cause discoloration and/or yellowing of the pipette housing. However, this will not affect the performance of the device in any way.

Regardless of the listed safety notes, additional applicable regulations and guidelines of trade associations, health authorities, trade supervisory offices, etc. must be observed.

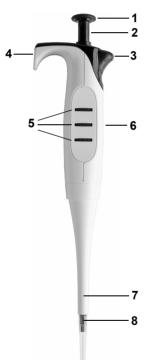
Please visit our website <u>www.integra-biosciences.com</u> on a regular basis for up to date information regarding REACH classified chemicals contained in our products.

2 Description of the device

2.1 Scope of delivery

- · EVOLVE manual pipette
- · Multipurpose tool
- · Color coded plunger cap
- · Quick start guide
- Replacement O-rings (for 200 μl, 1000 μl and 5000 μl models)

2.2 Overview of the EVOLVE manual pipettes



- 1 Plunger cap
- 2 Plunger
- 3 Tip ejector
- 4 Finger hook
- 5 Volume adjustment dials
- 6 Volume window



The color of the volume indicator label matches GRIPTIP rack insert

- 7 Ejector sleeve
- 8 Tip fitting

3 Operation

3.1 Setting the volume



- 1) To set the desired pipetting volume press down the **plunger** (a) and rotate slightly clockwise to release the volume dials.
- 2) Spin the **adjustment dials** (b) until the desired volume is shown in the volume window.
- Slightly rotate and slowly release the plunger (a) to set the volume

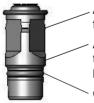
3.2 Attaching and removing GRIPTIP pipette tips



CAUTION

To ensure optimal performance of your EVOLVE manual pipettes always use GRIPTIPS of correct size which have been designated for the use with the EVOLVE manual pipettes, see "6.2 Consumables" on page 33.

The unique GRIPTIP system of the EVOLVE manual pipettes reduces attachment and ejection forces, ensures a perfect fitting to prevent the tips from falling off and provides a perfect seal. On a multichannel pipette, all tips sit at the same height.



A rim inside the GRIPTIPS snaps over the multi-lobes and ensures firm attachment of the tips.

A shoulder provides a positive stop to prevent over-tightening of the tips. Without hammering, the tip is either on or off but nothing in between.

O-ring provides forgiving and robust seal surface for the pipette tip.

Attach the tips:

When loading tip(s), press the pipette into the appropriate GRIPTIP(s) until you hear and feel a click indicating that a seal has been achieved. Once you feel the click, stop applying pressure. When loading GRIPTIP pipette tips using a multichannel pipette, slowly rock the pipette from one side to the other side to ensure the proper seal is achieved.

Discard your used tips:

If liquid is in the tips, empty them by pressing down the **plunger** ($\underline{2}$) to the second stop (see section $\underline{3.3}$). Tips are easily removed by pressing the **tip ejector** ($\underline{3}$).

3.3 Pipetting

The plunger can be pushed down to two different stops:



- 1) Top position: Varies depending on volume setting.
- 2) First stop: Fixed calibrated position for aspirating and dispensing.
- 3) Second Stop: To blowout the residual liquid.

For detailed recommendations for pipetting, see also <u>"3.3.3 Proper pipetting techniques"</u> on page 11.

3.3.1 Aspirating and dispensing

To aspirate and dispense liquid, perform the following steps:

Aspirate



- With GRIPTIPS attached, push down the plunger to the first stop.
- 2) Immerse the GRIPTIPS in liquid.
- 3) Slowly return **plunger** to the top position. For higher volumes (>200 µl), follow the liquid level by moving the pipette down.
- 4) Remove the GRIPTIPs from liquid.

Dispense



- 5) To dispense the liquid, push the **plunger** down to the first stop.
- 6) If there is any residual liquid in tip, a <u>blowout</u> should be performed by pressing the **plunger** to the second stop.
- 7) Remove the GRIPTIPS from vessel and release plunger.

3.3.2 Reverse mode

Viscous samples should be aspirated and dispensed at slower speeds and in reverse pipetting mode:

Aspirate



- 1) Fully push down the **plunger** to the second stop.
- Immerse the GRIPTIPS in the liquid, let the plunger slowly release to the upper position and wait shortly until the liquid is fully aspirated.

Dispense



- 3) To dispense, press the **plunger** slowly down to the first stop and wait a few seconds to dispense the liquid.
- 4) Remove the GRIPTIPS from the target vessel.
- 5) To expel the excess liquid, press the **plunger** to the second stop.

3.3.3 Proper pipetting techniques

INTEGRA Biosciences recommends the following techniques for enhancing pipetting results. These techniques are consistent with ISO standard 8655-2.

- It is best to immerse the GRIPTIPS just below the surface of the liquid (2–3 mm) to allow the desired volume to be aspirated. If the tip is immersed too deep, the risk of carry-over is increased due to liquid drops clinging to the outer surface of the tip.
- Press and release the plunger with consistent speed and smoothness.
- Always pre-wet GRIPTIPS. After loading tips onto your pipette, aspirate and dispense
 the full volume 2-3 times to coat the inside of pipet tips. Pre-wetting ensures that the
 liquid and air inside the tips are at equal temperature and the dead air space is humidified. Neglecting to perform a pre-wet can result in a smaller delivery volume in the first
 few dispenses.
- EVOLVE manual pipettes are air displacement pipettes. To properly dispense liquids, ensure that the pipette tip is at a 0–20° angle. After each dispense, you must touch GRIPTIPS against the vessel wall or liquid. This process is referred to as "touching off" or "tip touch" and prevents liquid from clinging to the pipette tips.
- Air displacement pipettes show the best performance between 35% and 100% of the nominal volume.
- Viscous samples should be aspirated and dispensed at slow speeds to ensure accurate pipetting. In addition, pipetting should be performed in a reverse mode.
- For pipetting liquids with high vapor pressures (such as methanol or ethanol), use relatively fast pipetting speeds and avoid prolonged pauses after aspiration.
- Calibrate based on fluid type. EVOLVE manual pipettes are tested and calibrated at
 the factory for use with distilled water at room temperature. It may be necessary to recalibrate your EVOLVE manual pipettes if the liquid to be used has different physical
 properties (specific gravity and vapor pressure) than water.



WARNING

Avoid pipetting for extended periods. To minimize the risk of repetitive strain injury, include pauses of several minutes.

Use filtered GRIPTIPS to pipette corrosive or biohazardous liquids. The filter prevents vapors and aerosols from corroding or contaminating the cylinder assembly.

3.4 Troubleshooting

Problem	Probable cause	Remedy
Leaking.	Tip incorrectly attached.Foreign particles between tip and tip fitting.	Attach a new tip.Clean tip fitting. Attach new tips
	Damaged colored O-ring.	 Change the O-ring, see <u>4.4.2</u>. If leak persists, contact service.
Dispense results are inaccurate.	 Unsuitable calibration for liquid class. Improper pipetting techniques. 	 Recalibrate with the liquids in question. Adjust aspiration and dispense speed depending on liquid: High viscosity liquids may require calibration. High vapor pressure liquids may require pre-wetting. Refer to proper pipetting techniques section 3.3.3
Not dispensing/ aspirating.	Piston stuck or not connected.Internal seal is damaged.	Contact service.
Droplets on the tips.	 Temperature of liquid differs from that of air inside the tips. Liquid of low viscosity and high vapor pressure. Touch off was not performed. 	Increase dispensing speed.

4 Maintenance

4.1 Cleaning

The materials used on the exterior of the EVOLVE manual pipettes support regular cleaning intervals. Clean the external components with a lint-free cloth lightly soaked with mild soap solution in distilled water or with a 70 % dilution of Isopropyl or Ethanol. Never use acetone or other solvents.



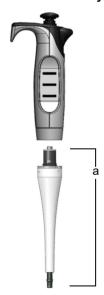
WARNING

- DO NOT immerse the entire pipette into the cleaning solution.
- DO NOT spray cleaning solution directly onto the exterior body of the pipette.

If the pipette is exposed to corrosive vapors, e.g. from concentrated acids, or if liquid enters the lower pipetting end, disassemble the lower part of the pipette and clean the internal parts with distilled water or with a 70 % dilution of Isopropyl or Ethanol. Follow the instructions below for disassembly.

4.2 Single channel lower end disassembly

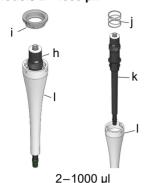
4.2.1 Disassembly



Models of all sizes:

- 1) Before you separate the lower part from a single channel pipette, press down the plunger.
- 2) Unscrew the **cylinder assembly** (a) clockwise and gently remove it from the pipette body.

Models 2-1000 µl:



Models 2-1000 µl:

3) With the cylinder assembly (a) in a hand, turn counterclockwise to remove the snap ring (i). Pull out black cylinder (k) from white sleeve (I). Remove the ejector spring (j).

Set the **snap ring** (i), **sleeve** (l) and **ejector spring** (j) aside or place them in an autoclave pouch.



Models 2 and 10 µl:

4) Unscrew **tip fitting** (e) from the **cylinder body** (k). Slide the **O-ring** (g) and the white **seal** (f) from the **piston** (h). If these parts are still stuck in the **tip fitting** (e), tap the top face of the tip fitting on the table.

Set these components aside or place them in an autoclave pouch.



Models 20 µl and 100 µl:

4) Unscrew tip fitting (e) from the cylinder body (k). Slide the O-ring (g) from the piston (h). If the O-ring (g) is still stuck in the tip fitting (e) after it has been removed, tap the top face of the tip fitting on the table.

Set these components aside or place them in an autoclave pouch.



Models 2 -100 μl:

5) Remove **self-containment cap** (p) from the **cylinder body** (k).

Pull out **piston** (h) from the **cylinder body** (k). Remove the **compression spring** (o) from the **piston** (d).

Set these components aside or place them in an autoclave pouch.



Model 200 µl:

5) Remove **self-containment cap** (p) from the **cylinder body** (k).

Pull out **piston** (h) from the **cylinder body** (k). Remove the **compression spring** (o) from the **piston** (h).

Set these components aside or place them in an autoclave pouch.



Model 1000 µl:

5) Remove cylinder adapter (q) from the cylinder body (k).

Pull out **piston** (h) from the **cylinder body** (k). Compress the **compression spring** (o) with one hand, then remove the **cup seal** (m) from **piston** (h).

Remove the **compression spring** (o) from the **piston** (h).

Set these components aside or place them in an autoclave pouch.

Model 5000 µl:



Model 5000 µl:

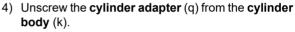
 Remove white sleeve cap (n) from white ejector sleeve (l).

Pull out **cylinder assembly** (a) from **ejector sleeve** (l).

Pull out **ejector spring** (j) from **cylinder assembly** (a).

Set the **sleeve cap** (n), **ejector sleeve** (l) and **ejector spring** (j) aside or place them in an autoclave pouch.

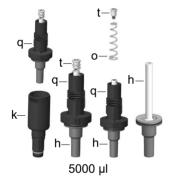
Model 5000 µl: 4) Unscrew the



Unscrew the **piston head** (t) from **piston assembly** (h).

Remove **compression spring** (o) from **piston** (h). Remove **cylinder adapter** (q) from **piston** (h).

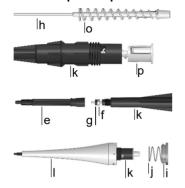
Set these components aside or place them in an autoclave pouch.



4.2.2 Reassembly

Before reassembling the pipette, check each component for lint or dust particles. It is recommended to replace the **O-ring** and **seal** or **cup seal**. Lightly lubricate the piston, O-rings and seals (see <u>"4.4.3 Lubrication" on page 21</u>).

Models 2 μl-100 μl:



- Slide the compression spring (o) into the piston (h).
 Place piston (h) into cylinder (k). Press the self-containment cap (p) into the cylinder (k), and
- 2) Slide the **seal** [(f); 2-10 µl only] and **O-ring** (g) into the **piston** (h) and lightly lubricate. Screw **tip fitting** (e) into **cylinder body** (k).

make sure it aligns with notches on cylinder (k).

3) Place the cylinder (k) inside the white sleeve (I). Place the ejector spring (j) over the cylinder (k). Place the snap ring (i) on the white sleeve (I). Turn the sleeve (I) clockwise until the snap ring (i) snaps into place and secures the cylinder in the cylinder assembly.

Model 200 µl:







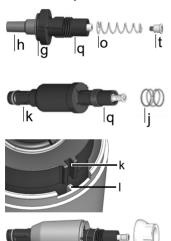
- Slide the cup seal (m), and the compression spring (o) into the piston (h), lightly lubricate cup seal (m). Be sure the compression spring (o) is placed into the piston (h) with the small end first.
- 2) Place the piston (h) inside the cylinder (k), and place self-containment cap (p) into the cylinder. Make sure self-contained cap (p) aligns with notches on cylinder.
- 3) Slide the **cylinder** (k) into the white **sleeve** (l). Place the **ejector spring** (j) over **cylinder**. Place **snap ring** (i) on the white **sleeve** (l). Turn the **sleeve** clockwise until the **snap ring** (i) snaps into place.

Model 1000 µl:



- Slide compression spring (o) into the piston (h).
 While compressing the spring (o) back, insert cup seal (m) into the piston (h) and lightly lubricate.
- Insert the piston (h) into the cylinder (k). Place cylinder adapter (q) into the cylinder (k). Make sure cylinder adapter (q) aligns with the pair of slots on cylinder.
- 3) Place **cylinder** (k) inside white **sleeve** (l). Place **ejector spring** (j) over cylinder. Place **snap ring** (i) on the white **sleeve** (l). Turn the **sleeve** clockwise until the **snap ring** (i) snaps into place.

Model 5000 µl:



- Place the cylinder adapter (q) over the piston rod (h). Slide the compression spring (o) into the piston (h). Screw piston head (t) into piston to secure the compression spring (o) and cylinder adapter (q). Lightly lubricate O-ring (g).
- Screw the piston assembly (h) to the cylinder (k).
 Slide the ejector spring (j) on the cylinder adapter (q).
- Place the cylinder (k) inside the white sleeve (I).
 Ensure to align the slots on the cylinder (k) with the bosses inside the sleeve (I).
- 4) Re-attach the **sleeve cap** (n) to the white **sleeve** (l). Ensure that the **sleeve cap** (n) aligns to the openings of the **sleeve** (l).

Models of all sizes:

- 5) Position the **piston** to the **piston drive plug** to connect the piston to the upper part of the pipette.
- 6) Screw the **cylinder assembly** (a) counterclockwise to attach it to the body of the pipette.

Perform a leak test (see <u>"4.5.5 Leak test" on page 24</u>) and validate pipetting volumes after reassembly.

4.3 Sterilization

If the surface of the EVOLVE manual pipettes have been in contact with biohazardous material, they must be decontaminated in accordance to good laboratory practice. Wipe the clean surface with a lint-free cloth, lightly soaked e. g. with the following disinfectants:

- Ethanol 70 %
- Microcide SQ 1:64
- Glutaraldehyde solution 4 %
- Virkon solution 1-3%

Follow the instructions provided with the disinfectants.

The device may be decontaminated with ${\rm H_2O_2}$ gas (maximal concentration 35 %) for 60 minutes.

It is not recommended to autoclave the EVOLVE manual pipettes. If autoclaving is required, only the lower assembly of the EVOLVE single channel pipettes can be autoclaved.



WARNING

Service is required after autoclaving the EVOLVE manual pipettes!

Do not autoclave the entire unit. Multichannel pipettes CANNOT be autoclaved, they might be damaged!

As-found calibration (measurement report, indicating "before" data) is not possible after autoclaving!

4.3.1 Autoclaving the disassembled components

Place the disassembled components (see $\underline{4.2.1}$) into steam autoclave in an autoclave pouch.

You may autoclave the components at 121°C, 1 bar overpressure for 20 minutes.



CAUTION

Service is required after autoclaving the EVOLVE Manual Pipettes!

4.4 Servicing

4.4.1 Shipping to INTEGRA Biosciences

For any service or repairs, please contact your local service technician.



WARNING

If working with infectious materials, e. g. human pathogens, the EVOLVE manual pipettes need to be decontaminated before sending to service and the declaration on the absence of health hazards must be signed. This is necessary to protect service personnel.

4.4.2 Changing O-rings of tip fittings

200 µl, 1000 µl and 5000 µl EVOLVE manual pipettes feature tip fittings with colored Orings. This O-ring is used to seal against the inside wall of GRIPTIPS and provides a robust seal.

O-rings are made of durable silicone. If necessary, e.g. in case of a leakage due to damaged O-ring, you can replace these O-rings. A set of spare O-rings can be ordered separately, see "6.2 Consumables" on page 33.



WARNING

Avoid mechanical damage of the tip fittings.



For 200 μ l or 1000 μ l pipettes, slide the multipurpose tool sideways onto the tip fitting until the O-ring (a) builds a loop. Remove the O-ring with plastic tweezers.

For 5000 µl pipettes, simply use plastic tweezers to remove the Oring.



Slide a new O-ring over the tip fitting (b).

4.4.3 Lubrication

The internal seals and O-rings are subject to wear. An undamaged thin lubricant film is important to keep the seals tight. The lubricant recommended depends on the EVOLVE pipette size, see <u>"6.2 Consumables" on page 33</u>:



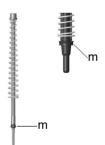
Single channel 2 µl - 10 µl models:

Use the Kluberalfa lube (#500-00182-00) to lightly lubricate the piston (h). Lightly lubricate outer ring of the **O-ring** (g) and the **white seal** (f).



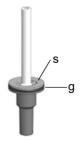
Single channel 20 µl and 100 µl model:

Use the Kluberalfa Lube (#500-00182-00) to lightly lubricate the **piston** (h). Lightly lubricate the outer ring of the **O-ring** (g).



Single channel 200 µl and 1000 µl models:

Use the Kluberalfa lube (#500-00182-00) to lightly lubricate the outer ring of the **cup seal** (m). Do not lubricate the bottom of **cup seal**.



Single channel 5000 µl model:

Use the Kluberalfa Lube (#500-00182-00) to lightly lubricate the outer ring of the **compression cap** (s) and **O-ring** (g). Do not lubricate the bottom of **compression cap** (s).

4.5 Calibration

The EVOLVE manual pipettes are factory tested and calibrated under environmentally controlled conditions using a gravimetric procedure in accordance with ISO 8655 standards. For the accuracy and precision specifications, see <u>"5.2 Pipette specifications"</u> on page 30.

For information regarding calibration service, please contact your local dealer.

4.5.1 Materials

- Precision balance with 0.01 mg readability
- · ASTM Class 2 or OIML E2 test weights
- Evaporation trap for balance
- Weighing vessel (optimally the height-to-diameter ratio is at least 3:1)
- · Measurement equipment for temperature, humidity and atmospheric pressure
- Distilled water (Grade 3 according to ISO 3696)
- New non-filtered GRIPTIPS matching the pipette volume (see section 6.2)
- · 1 beaker with distilled water

4.5.2 Definitions

Blow-out: Discharges any residual liquid from the pipette tip.

Blow-in: After the blow-out, the piston moves back into home position. This causes a slight intake of air (or liquid if tips remain in the liquid).

Two step blow-out: To avoid an early blow-in, keep the plunger pressed after dispensing, remove the tips from liquid and then release the plunger.

Pre-wet: The action of pre-coating the inside of the liquid contacting parts with a thin film of the same liquid. Additionally, it equilibrates humidity of the air space inside tip and the pipette.

Touch off: Touching the pipette tip against the surface liquid or side of well-plate to release any sample liquids that might be on the end of the pipette tip.

4.5.3 Test conditions and environment

Tests and calibrations should be performed in conditions and environment according to ISO 8655-6 standard.

- Temperature needs to be between 18–25 °C and remain constant (±0.5 °C) throughout the calibration.
- Optimal relative humidity of the environment is >50% and around the dispensing position 80%.
- GRIPTIPS, pipettes and distilled water need to be in the calibration laboratory for at least 2 hours prior to calibration to reach temperature equilibrium with the environment
- The balance must be validated using reference weights before and after a measuring series. A low and high volume weight should be used. E.g.:
 - 100 g (Mettler Toledo, #11119250)
 - 10 g (Mettler Toledo, #11119220)
 - 1 g (Mettler Toledo, #11119190)
 - 10 mg (Mettler Toledo, #11119130)



CAUTION

Always use new, pre-wetted GRIPTIPS for leak test and calibration.

4.5.4 Pre-wetting of tips

Pre-wet new GRIPTIPS three times prior to starting tests and calibrations. This is required every time a tip is changed.

- 1) Set the pipette to aspirate full volume.
- 2) Attach a new unused GRIPTIP which correspond to the volume range of the pipette directly from the rack without touching the tip by hand.
- 3) Use a separate waste container for pre-wet dispenses.
- 4) Push down the plunger to the first stop. Aspirate by releasing with medium speed.
- 5) Dispense: Keep the plunger pressed to the first stop until all liquid is expelled.

At the end of the third cycle push the plunger to the second stop, remove the tip from the liquid and then release the plunger to perform a two-step blowout.

4.5.5 Leak test

It is recommended to perform a leak test every 3 months or when errors occur.

- 1) Prewet tips as described above.
- 2) Set the EVOLVE manual pipettes to aspirate full volume.
- Push down the plunger to the first stop. Aspirate full volume by releasing with medium speed and verify liquid level is not decreasing during 10 seconds while GRIPTIPS are still in distilled water.
- 4) Remove the GRIPTIPS from the liquid and hold the pipette at a 20° angle. Wait for 20 seconds
 - a) Observe whether liquid droplets are forming at the end of the GRIPTIPS.
 - b) On a multichannel pipette, verify liquid level is even across all channels.
- 5) Immerse GRIPTIPS approximately 2 mm into distilled water and push and release the plunger to mix six times.
 - a) Observe whether air bubbles are forming when dispensing.
 - b) On a multichannel pipette, check whether the levels stay approximately on the same level across all channels.
- 6) At the end push the plunger to the second stop and remove the tip from the liquid before releasing.

Signs indicating a leak

1) During the mix cycle in the leak test, the liquid level of 1 channel is decreasing.



NOTE

A decreasing liquid level at aspiration could be an indication of a slow leak. Performing a retest at 10 mixes may help identify a slow leak.

- 2) Liquid is left in a tip after the last dispense during the leak test.
- 3) One or more channels show air bubbles during the mix cycle in the leak test.
- Droplets are forming when holding the pipette in the air for 20 seconds, even if a prewet was performed.
- 5) The liquid levels are not equal on all channels after aspiration.

If the pipette is leaking, change O-rings (see $\underline{4.4.2}$) and lubricate pistons (see $\underline{4.4.3}$) or contact your service technician.

4.5.6 Obtaining the actual volume

INTEGRA certifies the pipettes at 10% and 100% of the nominal value. For each volume 5 measurements are taken from all channels.

General

- 1) Always use new, unused GRIPTIPS, also when changing the test volume (e.g. from 200 µl to 20 µl).
- 2) Always pre-wet (section <u>4.5.4</u>) when using a new GRIPTIP. For a low volume measurement, first set the maximum volume for the pre-wet and then change to the volume to be measured.
- 3) After dispensing, perform a touch off to make sure no liquid remains on the tip.

Gravimetric testing

- 1) Write down the ambient temperature and air pressure.
- 2) Set the pipette to the high test volume.
- 3) Perform a pre-wet, see 4.5.4.
- 4) The first and second dispense to the balance should not be recorded. After each dispense re-tare the balance.
- 5) Aspirate the Target volume of water keeping the pipette in a vertical to 20-degree position while immersing the pipet tip 2–3 mm below the surface of the water. When withdrawing the tip from the liquid, gently wipe the tip against the side wall of the vessel to remove any liquid from the outside of the pipet tip.
- 6) Start the first measurement. Always pipet directly into the liquid of the weigh container on the balance. At the end push the plunger to the second stop and hold the plunger pressed until the pipette is removed from the weigh vessel to perform a 2-step blowout. Record the weight from the balance.
- 7) After completing the 5 high volume measurements, continue with the mid and low test measurements by repeating steps 3–6.

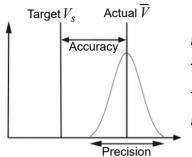
4.5.7 Calculation of accuracy and precision

Accuracy

The ability of a pipette to aspirate or dispense the exact volume desired. It indicates the proximity of measurement results to the true value. Accuracy is also known as systematic error and as the name indicates, can be corrected/calibrated.

Precision, coefficient of variation (CV)

Precision indicates the repeatability or reproducibility of the measurement. It is also called random error and is therefore an unpredictable error, which cannot be corrected/calibrated.



The following symbols are used throughout this text:

 $V_{\rm s}$ = Selected test or target Volume

 m_i = Measured Liquid Weight (g)

Z = Z factor, see 5.35.3

 V_i = Converted Volume (ml or μ l)

 \overline{V} = Actual mean Volume (ml or µl)

n = Number of measurements

Conversion of the mass to volume

The values obtained by balance readings are in grams. These values need to be converted to micro liters using the Z correction factor. It takes into account the water density and air buoyancy during weighing at the corresponding test temperature. To determine the correct Z factor, find the intersection between temperature and air pressure in Table $\underline{5.35.3}$. Round up temperature and air pressure values.

Multiply each weight m_i obtained in <u>4.5.6</u> with the corresponding Z factor:

$$V_i = m_i \times Z$$

Add together the volumes V_i delivered, divide the sum by n (e.g. n = 5) to calculate the mean volume \overline{V} (in milliliters or micro liters) delivered at the test temperature, which is the Actual Volume:

$$\overline{V} = \frac{1}{n} \times \sum_{i=1}^{n} V_i$$

Calculation of systematic error (accuracy)

The systematic error e_s can be calculated using the following equation with V_s being the selected test volume:

$$e_s = \overline{V} - V_s$$

or in percent:

$$e_s = \frac{100 \times (\overline{V} - V_s)}{V_s}$$

Calculation of random error (precision %)

To calculate the random error as the repeatability standard deviation s_r , use the following equation:

$$s_r = \sqrt{\frac{\sum_{i=1}^{n} (V_i - \overline{V})^2}{n-1}}$$

The random error can also be expressed as a percentage, by the coefficient of variation ${\it CV}$, using equation:

$$CV = 100 \times \frac{s_r}{\overline{V}}$$

 $\underline{\rm If}$ the pipette is properly calibrated, the Target volume $V_{\rm S}$ should equal the Actual volume \overline{V} within the accuracy specifications of the pipette.

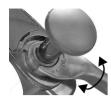
4.5.8 Adjusting EVOLVE manual pipettes

Compare the calculated accuracy and precision values with the corresponding pipette specifications given in section $\underline{5.2}$.

The industry standard is to test and present specifications using neat transfers. This is aspirating and dispensing the same volume.

In case an EVOLVE Manual Pipette has not met the calibration specifications it needs to be adjusted. This can be corrected with the Multipurpose Tool.

Put the two teeth of the Multipurpose Tool into the slots on the top of the pipette.



Rotate clockwise to increase the volume (slight turn up to a full turn). Rotate counterclockwise to decrease the volume (slight turn up to a full turn).



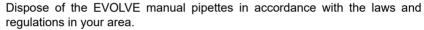
NOTE

Validate the new volume set by repeating the gravimetric testing.

4.6 Equipment disposal



The EVOLVE manual pipettes must not be disposed of with unsorted municipal waste.





In certain regions and countries, e.g. in EU member states, the distributor is obliged to take back this product free of charge at the end of life. Please contact your local distributor for more details.

5 Technical data

5.1 Environmental conditions

	Operation
Temperature range	5-40°C
Humidity range	Max. rel. humidity 80% for temperatures up to 31°C, decreasing linearly to 50% rel. humidity at 40°C.
Altitude range	<2000 m

5.2 Pipette specifications

The specifications apply only to neat transfers when the instrument is used together with INTEGRA GRIPTIPS and represent the performance of all channels on a multichannel pipette.

EVOL	VE manι	Manufactu	ırer			
Chan- nel	Part No.	Volume range (μl)	Volume increments (µI)	Test volume (µl)	Accuracy (±%)	Precision (≤%)
1	3011	0.2-2	0.001	0.2	12.0	6.0
				1	3.0	1.6
				2	1.5	0.7
1	3012	1–10	0.01	1	5.0	1.5
				5	1.5	0.8
				10	1.2	0.5
1	3013	2-20	0.01	2	7.0	2.0
				10	1.6	0.5
				20	1.0	0.35
1	3015	10-100	0.1	10	3.5	1.0
				50	1.0	0.35
				100	0.8	0.2
1	3016	20-200	0.1	20	2.5	1.0
				100	1.0	0.3
				200	0.7	0.18
1	3018	100-1000	1	100	3.0	0.6
				500	1.0	0.3
				1000	0.8	0.2
1	3019	500-5000	5	500	3.0	0.6
				2500	1.2	0.3
				5000	0.6	0.18

EVOL	VE manu	al pipettes			Manufactu	ırer
Chan- nel	Part No.	Volume range (µl)	Volume increments (µI)	Test volume (µl)	Accuracy (±%)	Precision (≤%)
8	3022	1–10	0.01	1	12.0	7.0
				5	4.0	3.0
				10	2.0	1.2
8	3024	5-50	0.05	5	5.0	2.0
				25	2.0	1.0
				50	1.2	0.5
8	3025	10-100	0.1	10	5.0	1.5
				50	2.0	8.0
				100	1.2	0.3
8	3026	20-200	0.1	20	5.0	1.2
				100	2.0	0.5
				200	1.2	0.3
8	3028	100-1250	1.0	125	5.0	1.1
				625	1.8	0.7
				1250	1.0	0.3
12	3032	1–10	0.01	1	12.0	7.0
				5	4.0	3.0
				10	2.0	1.2
12	3034	5-50	0.05	5	5.0	2.0
				25	2.0	1.0
				50	1.2	0.5
12	3035	10–100	0.1	10	5.0	1.5
				50	2.0	8.0
				100	1.2	0.3
12	3036	20-200	0.1	20	5.0	1.2
				100	2.0	0.5
				200	1.2	0.3
12	3038	100-1250	1.0	125	5.0	1.1
				625	1.8	0.7
				1250	1.0	0.3

EVOLVE manual pipettes					Manufacturer	
Chan- nel	Part No.	Volume range (µl)	Volume increments (µI)	Test volume (µl)	Accuracy (±%)	Precision (≤%)
16	3042	1–10	0.01	1	12.0	7.0
				5	4.0	3.0
				10	2.0	1.2
16	3044	5-50	0.05	5	5.0	2.0
				25	2.0	1.0
				50	1.2	0.5
16	3045	10-100	0.1	10	5.0	1.5
				50	2.0	8.0
				100	1.2	0.3

Precision = Coefficient of Variation.

5.3 Z correction factors

Temp.	Air pressure (kPa)						
(°C)	80	85	90	95	100	101.3	105
18.0	1.0022	1.0023	1.0023	1.0024	1.0025	1.0025	1.0025
18.5	1.0023	1.0024	1.0024	1.0025	1.0025	1.0026	1.0026
19.0	1.0024	1.0025	1.0025	1.0026	1.0026	1.0027	1.0027
19.5	1.0025	1.0026	1.0026	1.0027	1.0027	1.0028	1.0028
20.0	1.0026	1.0027	1.0027	1.0028	1.0028	1.0029	1.0029
20.5	1.0027	1.0028	1.0028	1.0029	1.0029	1.0030	1.0030
21.0	1.0028	1.0029	1.0029	1.0030	1.0031	1.0031	1.0031
21.5	1.0030	1.0030	1.0031	1.0031	1.0032	1.0032	1.0032
22.0	1.0031	1.0031	1.0032	1.0032	1.0033	1.0033	1.0033
22.5	1.0032	1.0032	1.0033	1.0033	1.0034	1.0034	1.0034
23.0	1.0033	1.0033	1.0034	1.0034	1.0035	1.0035	1.0036
23.5	1.0034	1.0035	1.0035	1.0036	1.0036	1.0036	1.0037
24.0	1.0035	1.0036	1.0036	1.0037	1.0037	1.0038	1.0038
24.5	1.0037	1.0037	1.0038	1.0038	1.0039	1.0039	1.0039

Z values in microliters per milligram

6 Accessories and Consumables

6.1 Accessories

Part No.
3200
3205
3210
3211
3214
3215

POPTOP base for reuse with ECO racks or GREEN CHOICE refills	Part No.
Small POPTOP base, for use with 12.5 µl, 125 µl and 300 µl GRIPTIPS	3250
Large POPTOP base, for use with 300 µl long and 1250 µl GRIPTIPS	3255

Color coded plunger caps (pack of 5)		Part No.
Black	for use with any pipette	3220
Purple	for use with 2 μl, 10 μl, 20 μl manual pipettes	3221
Yellow	for use with 50 μl, 100 μl manual pipettes	3222
Green	for use with 200 μl manual pipettes	3223
Blue	for use with 1000 μl manual pipettes	3224
Orange	for use with 5000 μl manual pipettes	3225

EVOLVE control spring assembly	Part No.
For single channel pipettes 2, 10, 20, 100, 200 and 1000 μ l only	3230

6.2 Consumables

O-rings	Part No.
for 200/300 µl tip fittings, pack of 24	100-00027-50
for 1000/1250 µl tip fittings, pack of 24	100-00028-50
for 5000 µl tip fittings, pack of 10	100-00029-00

Grease for pipettes and O-rings

Part No.

Kluberalfa lube, for single channel pipettes 20 μ l – 5000 μ l	500-00182-00
Nye fluorocarbon gel 807, 5 g, for single channel pipettes 2 μ l – 10 μ l	100-00136-50

6.3 **GRIPTIPS**

INTEGRA offers a wide range of GRIPTIPS in the volume ranges from 12.5 µl – 5000 µl.



Visit the GRIPTIP Selector Guide (www.integra-biosciences.com/griptips) to find the correct GRIPTIPS and set a filter by available volumes, packaging and properties.

6.3.1 **GRIPTIPS** for handheld pipettes

EVOLVE pipettes are compatible with GRIPTIPS for handheld pipettes.

6.3.2 Package options

- ECO Racks (3xxx series): lightweight PET racks, environmentally friendly with 60% less plastic. They have a carbon footprint half the amount of standard racks. Most convenient experience when paired with the reusable POPTOP Base.
- GREEN CHOICE (44xx series): environmentally friendly refills allowing the reuse of existing racks and thus reduces plastic waste.
- Standard racks (4xxx series): refillable with GREEN CHOICE inserts.
- · Bulk packs: tips in a resealable bag for hand loading.

6.3.3 **GRIPTIP** properties

According to our cleanroom standards, all GRIPTIPS (non-sterile, pre-sterilized and sterile) comply with our VIAPURE claims. This states that all product are RNase, DNase, Endotoxin and Pvrogenic free.

- · Sterile/pre-sterilized products are gamma irradiated within the minimum and maximum dosage range specified for INTEGRA sterile products. Racks are individually vacuum sealed in a bag and are considered sterile until opened. Pre-sterilized ECO Racks are individually latched and sealed with a heat shrink band. The entire case of 5 pre-sterilized GREEN CHOICE inserts are sealed.
- Non-sterile items are manufactured in the same cleanroom and packed in a carton. case.
- Long: longer design allows easy access into deep laboratory vessels
- Short: shorter design allows easy access in 1536 well plates or improves ergonomics
- Wide bore: large opening at the tip end, reduces shear forces
- Low retention: low liquid retention, for liquids with low surface tension

Imprint

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This operating instruction manual has part number 900-00010-00, the version is V07. It applies to EVOLVE manual pipettes as of serial number 2116000 or higher of until a newer revision is released.

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Manufacturer and customer service

Your local INTEGRA Biosciences representative, further information, and operating instructions in other languages can be found at www.integra-biosciences.com or are available on request info@integra-biosciences.com.

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