AXOL

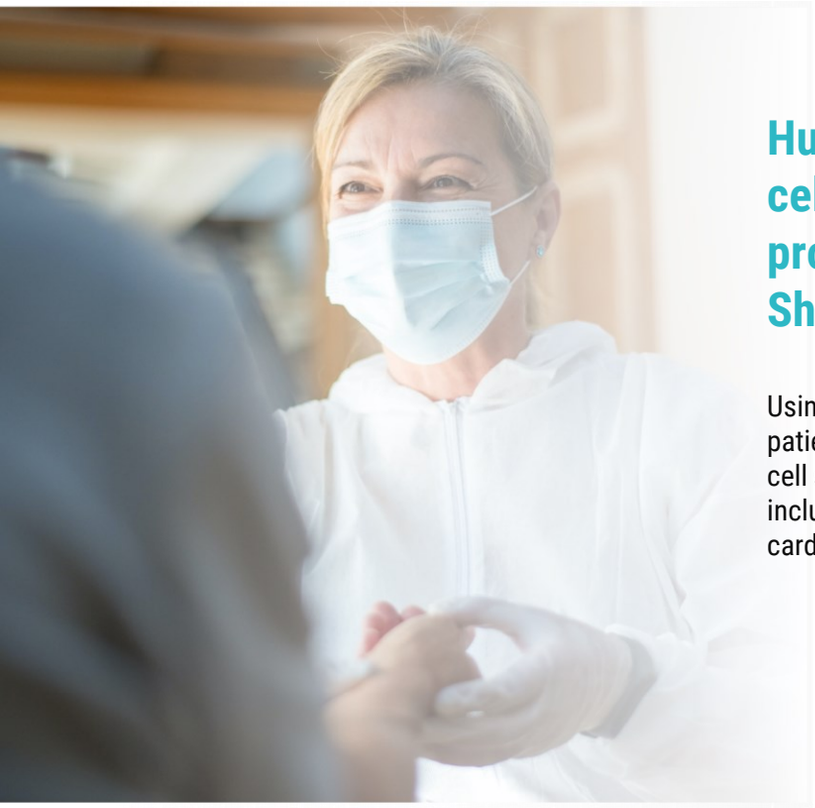
axoCells™

2024 Catalog

Functionally active iPSC-derived cells for drug discovery and research.

www.axolbio.com

Advanced *in vitro* systems.
From human donors to functional iPSC-derived cells for drug discovery and safety



Human induced pluripotent stem cells (human iPSCs) were first produced by Nobel Prize Winner Shinya Yamanaka in 2006.

Using this technology, consented blood or skin donations from patients and healthy donors are ‘reprogrammed’ into a stem cell state, from which they can be turned into any cell type including neurons, neuroinflammatory cells, muscle cells and cardiac cells.

Importantly, they retain behaviors of the disease state.

iPSC-derived cells can then be grown on their own (‘monocultures) or in mixtures ('co-cultures') of different cell types (for example, cortical excitatory neurons, inhibitory interneurons and neuroinflammatory cells) to make advanced *in vitro* models for research, toxicity studies and drug discovery.

These models can be used to test compounds for safety and effectiveness.

High-quality iPSC-derived cells can be used to fuel robust, scalable *in vitro* human disease models to accelerate and de-risk drug discovery.

axoCells™

With over a decade of experience, we've developed the manufacturing capabilities to produce **high-quality, functional iPSC-derived cells** with excellent consistency.

Your research can benefit from our **quality-focused approach**, with a catalog of iPSC-derived neurons, neuroinflammatory cells, muscle cells and cardiomyocytes manufactured at our **ISO 9001:2015-accredited** production facility.

Our leading neuronal cell types include: **cortical excitatory neurons**, **striatal neurons**, **cortical inhibitory interneurons**, **sensory neurons** and **motor neurons**. We also provide high-quality neuroinflammatory cells (**microglia and astrocytes),** cardiac cells (**atrial** and **ventricular cardiomyocytes)** and muscle cells **(skeletal myotubes).**

The cells listed in this catalog are available **'ready-to-ship'** as well as **'made-to-order'. 'Ready-to-ship'** cells are already manufactured in our ISO 9001-accredited production facility, stored frozen and available to purchase along with specialist media and supplements. Cells are also available **'made-to-order'** as custom differentiation production runs from your lines or ours: contact us at operations@axolbio.com for details.

Frozen cells

+

Specialist media and supplements

Kit format – cells, media and supplements

Cells, media and supplements can be purchased separately or often together as a kit.

SPEED IS TIME, COST AND SUCCESS

axoCells are designed to be functionally assay ready, faster.

Neurons

		Assay ready in
Cortical excitatory neurons		20 days
Cortical inhibitory interneurons		20 days
Sensory neurons	Powered by Maximizer supplement for faster, more *in vivo*-like development	21 days
Striatal neurons		31 days
Motor neurons	Powered by Accelerator supplement for faster, more *in vivo-like* development.	10 days

Muscle cells

Myotubes	5 days

Neuroinflammatory cells

Microglia	7 days
Astrocytes	2 days

Cardiac cells

Ventricular cardiomyocytes	7 days
Atrial cardiomyocytes	7 days

Guide to axoCells cell types

At Axol Bioscience, our iPSC-derived cells fuel **advanced *in vitro* systems** for drug discovery and drug safety.

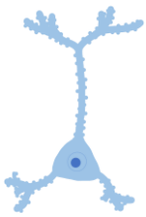
We provide functional cells to explore:

- **Neuroscience:** modeling neurodegenerative and neuroinflammatory diseases including ALS, Alzheimer's disease and Huntington's Disease
- **Pain and sensation:** modeling pain and sensory function for drug discovery and neurotoxicity testing
- **Cardiovascular:** using functional cardiomyocytes to model cardiac diseases (including arrhythmias) and for cardiotoxicity screening

Human disease	Cells commonly used to fuel *in vitro* models
Alzheimer's Disease (AD)	Cortical excitatory neurons, cortical inhibitory interneurons, microglia, astrocytes
Amyotrophic Lateral Sclerosis (ALS)	Motor neurons, skeletal myotubes, microglia
Huntington's Disease (HD)	Striatal neurons, cortical excitatory neurons, microglia, astrocytes

Neuroscience

Neurons



axoCells Cortical Excitatory Neurons

Cortical excitatory neurons are glutamatergic neurons that represent those found in the cerebral cortex.

- *Frequently used to fuel in vitro neuroscience models including AD, often used in co-culture models with other neuronal and neuroinflammatory cells. Supplied as neural stem cells (NSCs) with maturation to cortical excitatory neurons via our easy-to-follow protocol.*

axoCells Cortical Inhibitory Interneurons

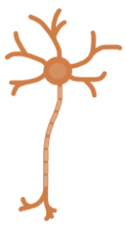
Cortical inhibitory interneurons are GABAergic neurons acting as the 'brakes' of the central nervous system. Connections between neurons in the brain are finely tuned and any increased electrical activity is dampened down by these cells.

- *Frequently used in co-culture methodologies for studying AD, seizure and epilepsy.*

axoCells Striatal Neurons

Striatal neurons represent neurons from the striatum, part of the basal ganglia which is related to movement control and reward. Striatal neurons progressively degenerate in patients with HD.

- *Frequently used in models of HD. Supplied as NSCs with maturation to striatal neurons via our easy-to-follow protocol.*



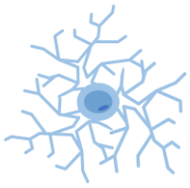
axoCells Motor Neurons

Motor neurons innervate muscle cells to control a range of voluntary and involuntary movements. The progressive destruction of motor neurons is associated with neuromuscular conditions including ALS.

- *Frequently used to fuel in vitro models of ALS with muscle cells (myotubes).*

Neuroscience

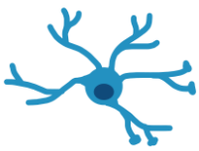
Neuroinflammatory cells



axoCells Microglia

Microglia are the key immune cells of the brain with crucial roles in brain development, neurogenesis, synaptic plasticity and homeostatic maintenance.

- *Used in co-culture with neurons for AD and ALS studies, and in monoculture for compound screening.*



axoCells Astrocytes

Astrocytes are a subtype of glial (supportive) cells and play critical roles in the regulation of blood flow, synapse function, and synaptic remodeling.

- *Used in co-culture with neurons for AD and ALS studies.*

Muscle cells

axoCells Myotubes

Human iPSC-derived myotubes can be matured into skeletal muscle cells for use in advanced *in vitro* musculoskeletal and neuromuscular model systems. Note, only available on custom 'made-to-order' service.

- *Used in neuromuscular research including ALS and models of the neuromuscular junction.*

Pain & Sensation

Neurons

axoCells Sensory Neurons

Sensory neurons are the nerve cells activated by sensory input from the environment, including touch, heat and pain.

- *Used in cosmetic sensitization testing and pain/touch models, often in microfluidic devices.*

Cardiovascular

Cardiac cells

axoCells Ventricular Cardiomyocytes

These ventricular cardiomyocytes represent those found in the human ventricles and are used to fuel *in vitro* cardiotoxicity and cardiac models to assess drug safety.

- *Used to fuel in vitro cardiotoxicity assays and for cardiac research.*

axoCells Atrial Cardiomyocytes

These atrial cardiomyocytes represent those found in the human atrium. They have been developed to support testing of irregular and abnormally fast heart rates (including atrial fibrillation),

- *Used for in vitro cardiotoxicity assays and for cardiac arrhythmia research.*

How to access axoCells

axoCells are available through two production routes:

‘Ready-to-ship’ axoCells

Already manufactured in our ISO 9001-accredited production facility and ready to ship. Stored frozen and available to purchase along with specialist media and supplements.

'Made-to-order’ axoCells

Custom differentiation production run from our lines or your lines, minimum order quantity 10 vials.

	axoCells kit code	iPSC-derived cells only	Donor	Disease	Gene/ mutation
Neurons					
Cortical Excitatory Neurons	ax5111	ax0111	Female, 87	Alzheimer's disease	Apo E4/E4
	ax5112	ax0112	Female, 38	Alzheimer's disease	PSEN1 L286V
	ax5113	ax0113	Male, 53	Alzheimer's disease	PSEN1 M146L
	ax5114	ax0114	Female, 31	Alzheimer's disease	PSEN1 A246E
	ax5115	ax0015	Male, newborn	Healthy control	-
	ax5116	ax0016	Female, newborn	Healthy control	-
	ax5118	ax0018	Male, 74	Healthy control	-
Striatal neurons	ax3115	ax0015	Male, newborn	Healthy control	-
	ax3116	ax0016	Female, newborn	Healthy control	-
	ax3118	ax0018	Male, 74	Healthy control	-
	ax3211	ax0211	Female, 48	Huntington’s disease	HTT >50 CAG
Cortical Inhibitory Interneurons	-	ax0662	Male, 40-50	Healthy control	-
	-	ax0667	Male, newborn	Healthy control	-
					-
Sensory Neurons		ax0555	Male, newborn	Healthy control	-
	ax0157	ax0055	Male, newborn	Healthy control	-
Motor Neurons		ax0073	Male, 62	ALS control (asymptomatic)	C9ORF72 >145 G4C2
		ax0074	Female, 64	ALS disease	C9ORF72 >145 G4C2
	ax0178	ax0078	Male, 74	Healthy control	-
Neuroinflammatory cells					
Microglia	ax0679	ax0664	Male, 40-50	Healthy control	-
Astrocytes	-	ax0665	Male, newborn	Healthy control	-
Cardiac cells					
Ventricular Cardiomyocytes	ax2500	ax2508	Male, 74	Healthy control	-
Atrial Cardiomyocytes	ax2510	ax2518	Male, 74	Healthy control	-
Cardiac Fibroblasts	-	ax3039	Adult	Healthy control	-

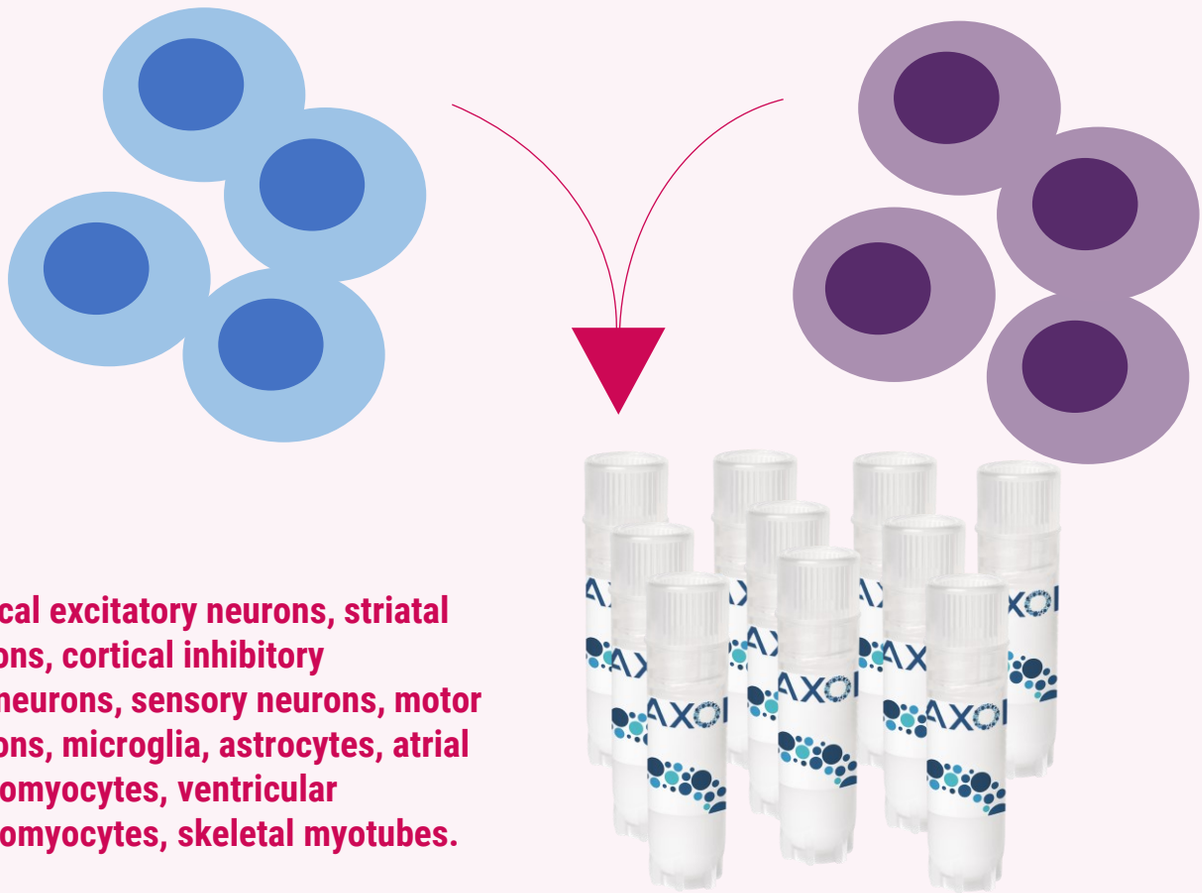
Custom Differentiation ‘Made-to-order’ **axoCells™**

We provide custom differentiation services, **‘made-to-order’** production runs, using iPSCs from our axoLines range or using your lines. There is a minimum order quantity of 10 vials. When using your lines, a review of ethics, quality and line onboarding will be required.

Custom Differentiation Runs
‘Made-to-order’ axoCells

Using iPSCs from our library of 70 axoLines as the starting material.

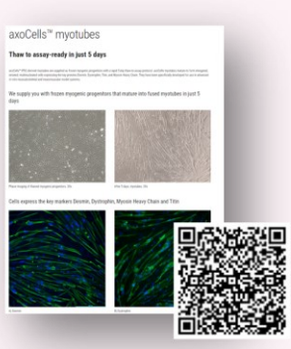
From your lines*.
*A review of ethics, quality and line onboarding will be required.



Cortical excitatory neurons, striatal neurons, cortical inhibitory interneurons, sensory neurons, motor neurons, microglia, astrocytes, atrial cardiomyocytes, ventricular cardiomyocytes, skeletal myotubes.

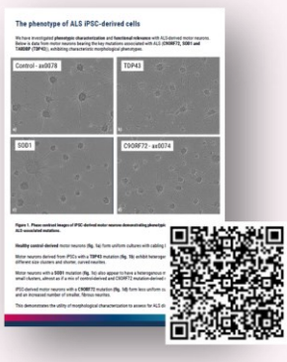
Minimum order quantity of made-to-orders is 10 vials.

Examples of ‘made-to-order’ cells in action:



Skeletal myotubes

TREM2 microglia



SOD1 and TDP43 motor neurons

axoLines™ – used to power the production of our cells

We have developed a library of over **70 iPSC lines** derived from fully-consented patient and healthy control donors. With **full licenses** and a **50:50 split of male to female donors**, our axoLines iPSCs are used as the basis for our cell manufacturing. Please note that axoLines iPSCs are not available for purchase.

Key therapy areas include **ALS, Alzheimer's Disease**, **Parkinson's Disease**, **Huntington's Disease**, **Friedreich's Ataxia** and **Frontotemporal Dementia**.

Key highlights from our axoLines range:

ALS lines

We have 5 iPSC lines reprogrammed from patients with ALS (**SOD1**, **TDP43** and **C9ORF72** mutations)

Examples include:

- *CENSOi035-B,* an iPSC reprogrammed from a 61-year-old female with ALS (**SOD1** mutation)
- *CENSOi018-A,* an iPSC reprogrammed from a 62-year-old female with ALS (**TDP43** mutation)
- We also have the interesting combination of a disease line derived from 64-year-old female with ALS (**C9ORF72** mutation) and a control line from their sibling, a 62-year-old male with a **C9ORF72** mutation who was **asymptomatic** at the time of sampling
- We also have a disease control line reprogrammed from a 44-year-old female with a **C9ORF72 mutation** (**asymptomatic** at time of sampling)

Alzheimer's Disease lines

We have 7 iPSC lines reprogrammed from patients with Alzheimer's Disease (**APOE4** and **PSEN1** mutations)

Examples include:

- An iPSC reprogrammed from an 87-year-old female with Alzheimer's Disease (**APOE4/E4** mutation)
- An iPSC reprogrammed from a 53-year-old male with Alzheimer's Disease (**PSEN M146L** mutation)
- For a control line, we recommend the *CENSOi004-E line* reprogrammed from a 40-50-year-old male

Parkinson's Disease lines

- We have 14 iPSC lines reprogrammed from patients with Parkinson's Disease representing mutations in **Ataxin-3**, **PINK1**, **PARKIN**, **PARK2**, **SNCA** and **LRRK2**

  Examples include:

  - *CENSOi028-A,* an iPSC reprogrammed from a 52-year-old female with Parkinson's Disease (**PINK1**)
  - *CENSOi030-A,* an iPSC reprogrammed from a 54-year-old female with Parkinson's Disease (**SNCA**)
  - For a control line, we recommend *CENSOi004-E* reprogrammed from a 40-50-year-old male

Huntington's Disease lines

- We have 6 iPSC lines reprogrammed from patients with Huntington's Disease representing mutations in **HTT**

  Examples include:

  - *CENSOi017-A,* an iPSC reprogrammed from a 51-year-old female with Huntington's Disease (**HTT**)
  - *CENSOi053-A,* a disease control line reprogrammed from a 64-year-old male who was **asymptomatic at the time of sampling** (**HTT**)

You can see the full list on our axoLines page at https://axolbio.com/axolines/ or by scanning the QR code.

Next-level quality manufacturing

For iPSC technology to fulfil its exciting potential, we believe that **quality**, **performance** and **consistency** must always be the priority. Over the last decade, we've invested heavily in our manufacturing scale and quality to produce consistent, functionally relevant cells. The key to this is our **ISO 9001-accredited production facility** in Roslin, Edinburgh, and our **quality-focused approach**.

When customers work with us, they benefit from our long-standing history of meeting and exceeding industry best practices.

Highlights of our quality manufacturing capabilities include:

- A manufacturing run QC success rate of **92%** in 2023
- An OTIF of **97%** in 2023 (well above the target of >93.5%)
- **100%** patient donor consent and licensing
- Batch runs of up to **250 x 1 million vials** of axoCells Microglia
- **49 products** manufactured in-house with **6 specialist cell-matched media**

Our ISO 9001-accredited production facility forms the bedrock for our global iPSC community, allowing us to reach over **1,300 customers** in **57 countries**, including **17 of the top 20 pharma companies.**

Demonstrating an ISSCR-compliant quality management system

The International Society for Stem Cell Research (ISSCR) Standards Document lays out gold-standard quality frameworks for stem cell researchers across 5 key areas:

- Basic characterization
- Pluripotency and the undifferentiated state
- Genomic characterization
- Stem cell-based model systems
- Reporting

Our scientists conducted a thorough analysis of our current processes against the ISSCR Standards Document, and we're delighted to report **excellent compliance** with the guidance.

Scan the QR code or visit our website to view the report.


Axol Bioscience response to ISSCR standards document 2023

AXOL

Axol Bioscience and the new ISSCR (International Symposium for Stem Cell Research) **Standards Document**

We are delighted to see the release of the new ISSCR Standards Document 2023.

Our quality and scientific team have reviewed the guidelines and are pleased to report on the following compliance statement.

*"There is a **strong history** through Roslin Cells' involvement with EBISC (European Bank for Induced pluripotent Stem Cells) and the UK Stem Cell Bank, in the early days of developing stem cells. We are **deeply rooted in compliance and quality**, having been part of the consortium setting the standards for quality-controlled stem cell lines. Several of us were involved in the **early foundational work** for these guidelines, so we are delighted to see the publication of the Standards Document along with **Axol's continued compliance** to these high standards."*

John Gardner
Manufacturing Operations Manager at Axol's Roslin site.

Committed to quality and consistency

Given the enormous potential for human iPSCs in building more human-relevant disease models, we prioritize the consistency and quality of our products. Our Quality Management System is the bedrock for our daily work, driving quality and consistency for every one of our customers.

Our Quality Management System

- Bulk / same batch manufacturing and storage are available
- Our manufacturing facilities are ISO 9001:2015 accredited
- We conduct in-depth iPSC characterization, both pre- and post-differentiation
- Our Certificate of Analysis inserts report on the quality testing methods and result
- We provide full batch control and options of ownership
- There is full traceability of material origin and source
- We utilize a Q-Pulse Quality Management System for compliant audit-trail operation

Standards for Human Stem Cell Use in Research

The ISSCR Standards Document 2023

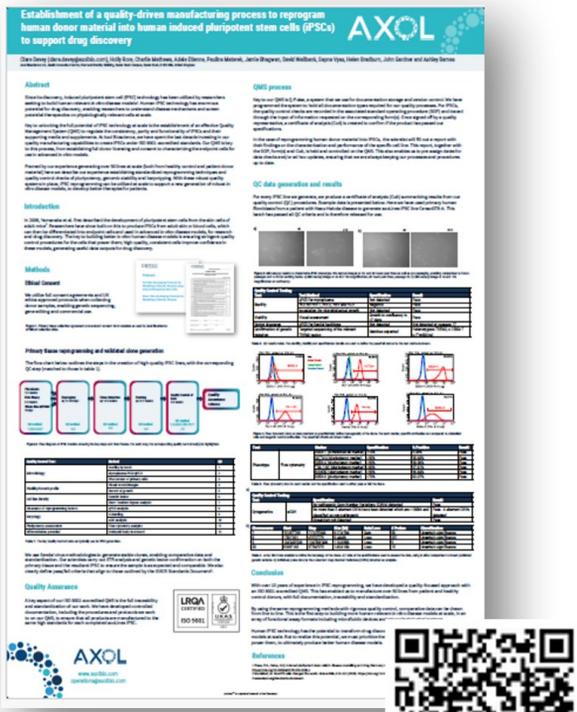
We have over a decade of experience supplying high-quality products and services to biopharma
We therefore welcome the publication of the ISSCR Standards Document 2023 and are pleased t
compliance with the individual components overleaf.

Establishing quality iPSC manufacturing procedures

Read more about our work exploring quality cell manufacturing processes in this poster: **"Establishment of a quality-driven manufacturing process to reprogram human donor material into human induced pluripotent stem cells (iPSCs) to support drug discovery".**

This work, presented at the Society for Laboratory Automation and Screening (SLAS) International Conference, explored the use of **standardized iPSC reprogramming methods** with **rigorous quality control** to enable line-to-line comparisons. We see this as the first step to building more human-relevant *in vitro* disease models at scale, in an array of functional assay formats including microfluidic devices and microphysiological systems.

Scan the QR code to access the poster.


Establishment of a quality-driven manufacturing process to reprogram human donor material into human induced pluripotent stem cells (iPSCs) to support drug discovery

AXOL

Abstract

Introduction

Methods

Ethical Consent

Primary tissue reprogramming and validated clone generation

Quality Assurance

QMS process

QC data generation and results

Conclusion

References

AXOL

Functional QC - a new step in the quality chain, building translational power into iPSC-derived models

We look to build *in vitro* models that mimic the human environment to support drug discovery programs. Existing quality control methods *characterize* cells, but we want confidence in whether they will actually *perform* in a model system- **ultimately, will these *in vitro* models be *useful* to researchers?**

At Axol Bioscience, we're looking to the subject of **functional quality control (fQC).** This is a new layer to our quality management system and will serve to **validate the functional performance** of our iPSC-derived axoCells within the context of an assay system.

+ QC + fQC = **Better human disease models**

How do we see functional QC working?

We're looking to add a new layer to our quality control, testing the **utility and performance** of cells in assays that are **biologically relevant** and **useful to research**. We believe this new standard will enhance **confidence** in the physiological relevance of our cells and drive **better translational power** in advanced *in vitro* models.

So as part of our cell manufacturing process, we will look to run cells within a 'real life' assay environment to assess performance within the model system on a batch-to-batch basis: only then will they pass fQC.

This fQC data will be built into our release packages and Certificate of Analysis statements.

What do you think?

We would like your thoughts on this, so please contact us at operations@axolbio.com

Scan the QR code to read the full discussion piece

Examples of fQC for iPSC–derived cells

axoCells Sensory Neurons are optimized for use in advanced *in vitro* pain models. fQC will measure the response to capsaicin at 1µM and 10µM concentrations via multi-electrode array (fig. 1).

We will also be looking to implement fQC for:

- **axoCells Motor Neurons** (measuring the appearance of synchronized burst firing via multi-electrode array) for ALS drug discovery
- **axoCells Microglia** (measuring bait uptake over 24 hours against a pre-determined threshold, with inhibition when adding cytochalasin D.) for neurodegenerative disease modeling

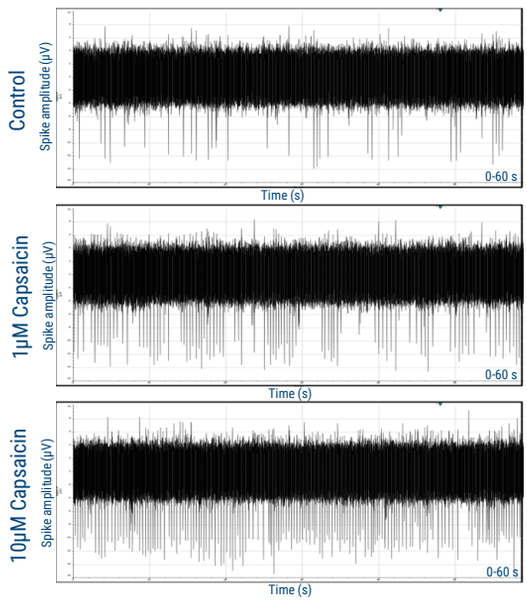


Figure 1. axoCells Sensory Neurons demonstrating response to 1µM and 10µM capsaicin at 22 days, measured on the Axion Maestro Pro multi-electrode array (MEA) system.

World leaders in manufacturing iPSCs consistently and at scale

1. Manufacturing performance

- Batch runs of up to **250 x 1 million** vials of axoCells Microglia
- **75** manufacturing events to date in 2023 and **118** in 2022
- **49** products manufactured in house
- 2023 manufacturing run QC success rate – **92%**
- **100%** patient donor consent and licensing
- **6** specialist cell matched media

2. Delivery excellence

- 2023 OTIF – **97%** (target >93.5%)
- Complaints rate **0.2%**
- **24/7** remote storage monitoring and offsite backup

3. Experience & technical expertise

- **16** years of experience
- **13** PhDs
- **>140** years of collective technical expertise in-house

4. Commercial reach

- Over **1300** customers worldwide
- Supplying to **57** countries
- Serving **17** of the top 20 pharma
- Supplying **OEM** and **MPS** platform integration customers

Delivering around the world

Our commitment to responsible shipping

At Axol Bioscience, our Shipping and Logistics division is central to our aims and we strive to ensure our global customers receive their product **on time and in full**, with open communication and a helpful approach. We pride ourselves on our **responsible shipping practices,** which means **communicating closely** with our customers and **designing shipping schedules** to maximize success- for example, shipping to the US on a Friday so it doesn't arrive over the weekend.

There are two ways in which we ship iPSC products:

- Cells and some media are shipped at -80°C in a polystyrene box containing dry ice
- Most of our media and some supplements are shipped on ice packs in polystyrene boxes at 2-8°C

Locally, we partner with our authorized **Distribution Partners**, your experts in technology, logistics and service to support your program of research. For more about your local distributor visit **axolbio.com/distributors/**

Need some extra support?

If you have any further questions or would like extra support, please get in contact at support@axolbio.com. We're here to help!

Scan the QR code to take a look at our **shipping infographic** and visit our website for handy shipping FAQs.

Neuroscience

Building better *in vitro* neurodegeneration and neurotoxicity models with human iPSC technology

Neuroscience

Better models for neurodegenerative disease and neurotoxicity

- Cortical excitatory neurons
- Striatal neurons
- Cortical inhibitory interneurons
- Microglia
- Astrocytes
- Myotubes
- Motor neurons

Neurodegenerative diseases (including **Alzheimer's Disease, Huntington's Disease and ALS)** are characterized by the **progressive and debilitating** loss of nervous function and control. They currently affect **tens of millions of patients** worldwide and, with our aging population and increasing co-morbidities, **rates of these conditions are rising.**

There is a **clear need for new therapies.** While animal models have provided valuable insights, researchers are looking to use more **human-relevant models** using **iPSCs** in a bid to close the **translational gap** that has seen new therapy failure rates as high as **99.6%.**

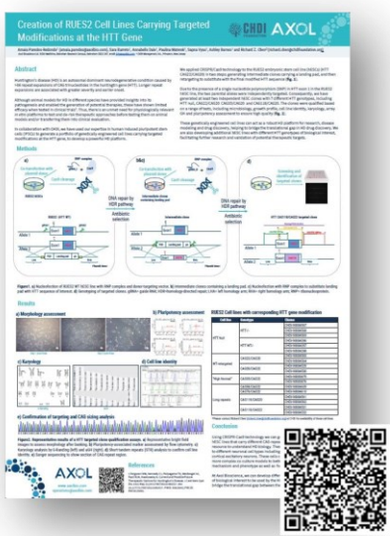
We have specifically developed a range of **axoCells iPSC-derived cells** for use in neurodegenerative disease models including ALS, Alzheimer's Disease and Huntington's Disease.

Key highlights include:

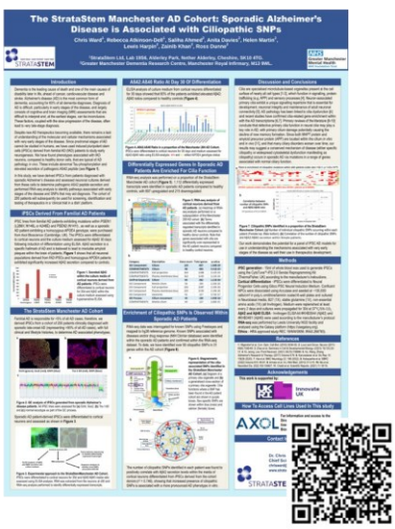
- **Phenotypic characterization**, including morphology and key marker expression
- **Functional performance** measured on a range of assays including electrophysiology, calcium imaging and neurite outgrowth
- **Designed for co-culture** and other advanced *in vitro* models including microfluidics and organ-on-chip devices
- **High-quality manufacturing** from our ISO 9001-accredited production facility

With a range of healthy control-derived and patient-derived cells, you can unlock the benefits of iPSC technology for your *in vitro* neurodegenerative disease and neurotoxicity models. Read more about our work in specific disease areas by **scanning the QR codes below:**

ALS



Huntington's Disease



Alzheimer's Disease

axoCells, designed with co-culture in mind

While monoculture models offer a valuable format for iPSC-based research and drug discovery, stepping up from monoculture to co-culture can unlock **better insights**, greater **human relevance** and **improved data outputs,** making co-culture models a valuable addition to *in vitro* research projects.

Co-culture models involve the use of **two or more iPSC-derived cell types** in an *in vitro* format. This can help researchers to incorporate **greater complexity**, **cell-cell crosstalk** and a more **physiological representation** of the *in vivo* environment.

As iPSC experts, we know that co-culture models can seem **technically challenging** to establish and execute. That's why we offer our expertise to unlock the benefits of iPSC-based co-culture models for neurodegenerative and cardiac researchers.

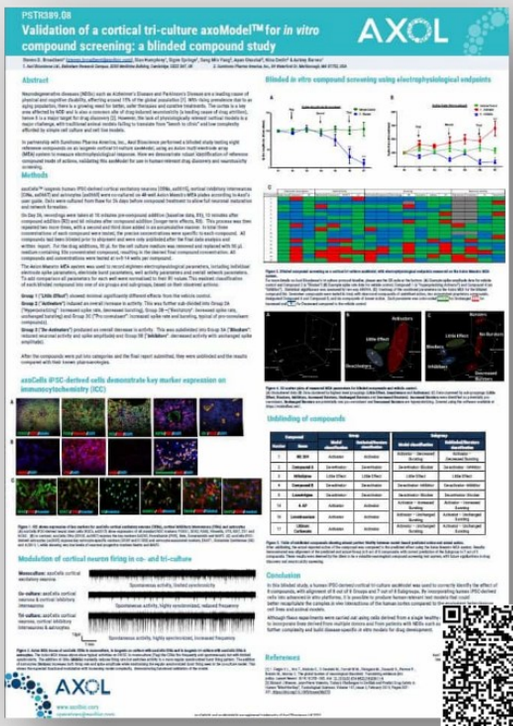
With single medias that support multiple cell types, we are building our cells with the future of co-culture in mind.

Validation of a cortical tri-culture axoModel™ for *in vitro* compound screening: a blinded compound study

With over a decade of iPSC expertise and experience building *in vitro* co-culture models, we've become the **first choice** for researchers looking to build *in vitro* co-culture models with multiple iPSC-derived cells.

Here is an example of an **axoModel system** built as a **complex tri-culture model** for compound screening. We incorporated axoCells Cortical Excitatory Neurons, cortical inhibitory interneurons and astrocytes to produce a cortical tri-culture model. This model was **successfully tested** against a blinded panel of compounds, demonstrating its value as a **compound screening platform** and paving the way for future testing of **neurotoxic liability**.

This work, produced in partnership with Sumitomo Pharma America Inc., demonstrates the **exciting potential** for advanced *in vitro* models in drug discovery and safety pharmacology, as well as our capabilities in building *in vitro* co-culture models. **Scan the QR code to access the poster.**


PSTR389.08

Validation of a cortical tri-culture axoModel™ for *in vitro* compound screening: a blinded compound study

AXOL

Abstract

Methods

Blinded in vitro compound screening using electrophysiological endpoints

axoCells iPSC-derived cells demonstrate key marker expression on immunocytochemistry (ICC)

Modulation of cortical neuron firing in co- and tri-culture

Unblinding of compounds

Conclusion

References

AXOL

axoCells™ Cortical Excitatory Neurons

axoCells Cortical Excitatory Neurons are **glutamatergic neurons** that represent those found in the human cerebral cortex. They are frequently used to fuel ***in vitro* neuroscience models** (including Alzheimer's Disease) in **co-culture** with other neuronal and neuroinflammatory cells. We supply them as neural stem cells (NSCs) with maturation to cortical excitatory neurons via our easy-to-follow protocol.

Assay ready in **20 days**

Express over 12 of the **key markers** (including FOXG1, PAX6 and vimentin)

Demonstrated **functionality** in advanced *in vitro* models including **co-culture and tri-culture**

Phenotypic characterization

We've **extensively characterized** our axoCells Cortical Excitatory Neurons to ensure the **correct morphology (fig. 1)** and expression of **key cell markers via immunocytochemistry (fig. 2).**

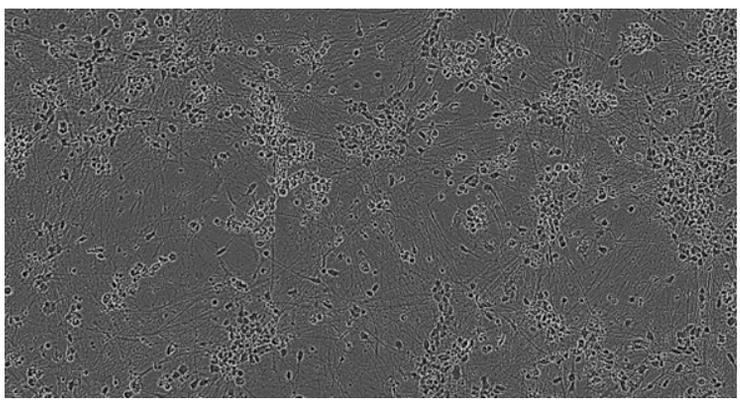
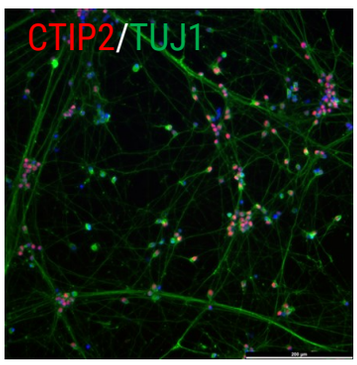


Figure 1. Phase image of axoCells Cortical Excitatory Neurons (ax0015) plated at 150,000 cells per cm^2 on day 20 of maturation. This demonstrates a pure population with regular morphology and neurite extensions. Image 20X.



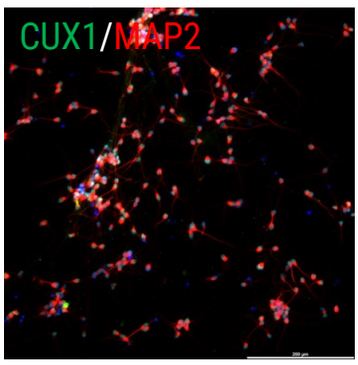


Figure 2. Immunocytochemistry of axoCells cortical neurons fixed at day 20 of maturation and stained for key markers CTIP2, TUJ1, CUX1 and MAP2.

Modulation of cortical neuron firing in co- and tri-culture

Monoculture: axoCells cortical excitatory neurons

Co-culture: axoCells cortical neurons & cortical inhibitory interneurons

Tri-culture: axoCells cortical neurons, cortical inhibitory interneurons & astrocytes

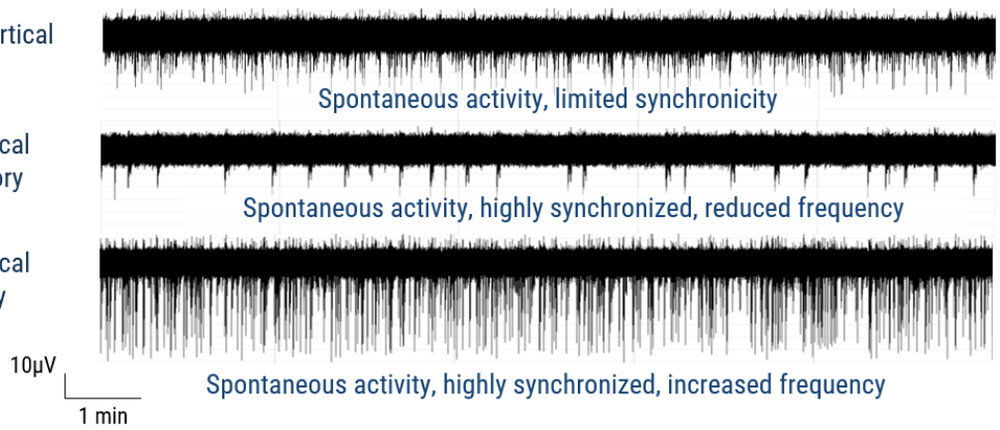


Figure 3. Axion MEA traces of axoCells Cortical Excitatory Neurons (CENs) in monoculture, in isogenic co-culture with axoCells Cortical Inhibitory Interneurons (CINs) and in isogenic tri-culture with axoCells CINs & astrocytes. The Axion MEA traces above show typical activities on DIV35. In **monoculture** the CENs fire frequently and spontaneously but with limited synchronicity. The addition of CINs (**co-culture**) markedly reduces firing rate but switches activity to a more regular synchronized burst firing pattern. The addition of astrocytes (**tri-culture**) increases both firing rate and spike amplitude while maintaining the regular synchronized burst firing seen in the co-culture model. This shows the expected functional modulation with increasing model complexity, demonstrating functional validation of the model.

Validation of a cortical tri-culture axoModel for *in vitro* compound screening a blinded compound study

With over a decade of iPSC expertise and experience building *in vitro* co-culture models, we've become the **first choice** for researchers looking to build *in vitro* co-culture models with multiple iPSC-derived cells.

Here is an example of an **axoModel system** built as a **complex tri-culture model** for compound screening. We incorporated axoCells Cortical Excitatory Neurons, cortical inhibitory interneurons and astrocytes to produce a cortical tri-culture model. This model was **successfully tested** against a blinded panel of compounds, demonstrating its value as a **compound screening platform** and paving the way for future testing of **neurotoxic liability**.

This work, produced in partnership with Sumitomo Pharma America Inc., demonstrates the **exciting potential** for advanced *in vitro* models in drug discovery and safety pharmacology, as well as our capabilities in building *in vitro* co-culture models.

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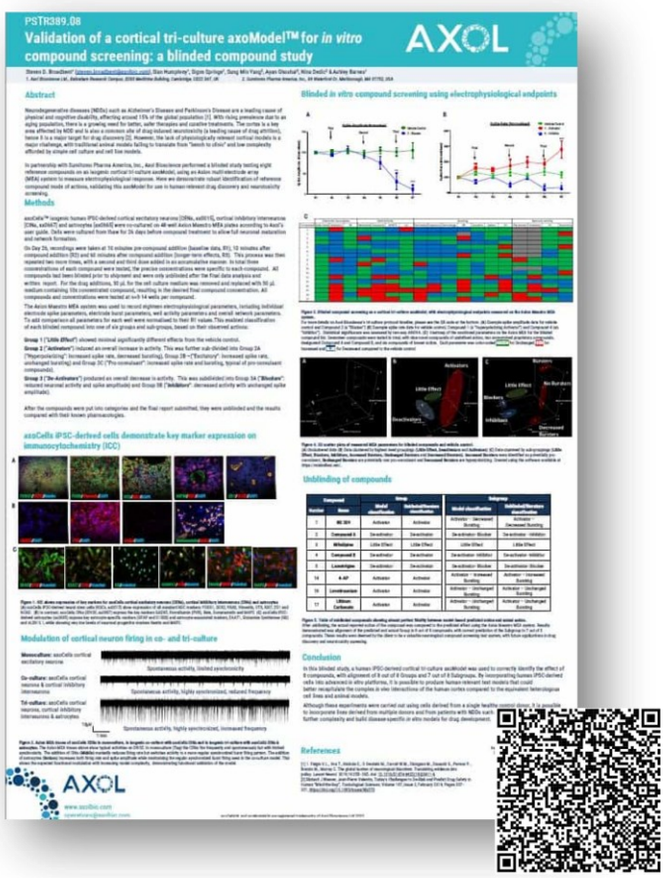


axoCells Human iPSC-Derived Cortical Excitatory Neurons

Supplied as Neural Stem Cells and a protocol to mature into final cortical excitatory neurons in **20 days**

Cells & Kits

Product Name	Cells only code / 1 vial	Quantity* / per vial	Kit** code
axoCells™ Human iPSC-Derived Neural Stem Cells, new-born male donor, ≥1.5 million cells	ax0015	≥1.5 million cells	**ax5115**
axoCells™ Human iPSC-Derived Neural Stem Cells, female donor, ≥1.5 million cells	ax0016	≥1.5 million cells	**ax5116**
axoCells™ Human iPSC-Derived Neural Stem Cells, male donor, ≥1.5 million cells	ax0018	≥1.5 million cells	**ax5118**
axoCells™ Human iPSC-Derived Neural Stem Cells, Alzheimer's Disease (APOE4 HOM) female donor, ≥1.5 million cells	ax0111	≥1.5 million cells	**ax5111**
axoCells™ Human iPSC-Derived Neural Stem Cells, Alzheimer's Disease (PSEN1 L286V) female donor, ≥1.5 million cells	ax0112	≥1.5 million cells	**ax5112**
axoCells™ Human iPSC-Derived Neural Stem Cells, Alzheimer's Disease (PSEN1 M146L) male donor, ≥1.5 million cells	ax0113	≥1.5 million cells	**ax5113**
axoCells™ Human iPSC-Derived Neural Stem Cells, Alzheimer's Disease (PSEN1 A246E) female donor, ≥1.5 million cells	ax0114	≥1.5 million cells	**ax5114**

*Number of viable cells post thaw
**Kit contains cells and one of each item listed in the media and supplements table

Media and Supplements

Product Name	Product code	Quantity
axoCells™ Neural Maintenance Media, 500ml	ax0031	500 ml
axoCells™ Human iPSC-Derived Cortical Neuron NeurOne Supplement	ax0674	1 ml + 1 ml
axoCells™ Human BDNF Supplement, 10 µg	ax139800	10 µg
axoCells™ Human GDNF Supplement, 10 µg	ax139855	10 µg
axoCells™ Neural Plating Media, 30ml	ax0033	30 ml
axoCells™ SureBond-XF Coating, 1 ml	ax0053	1 ml

Additional third-party components may be required. Please refer to protocol for full list.

User Protocol

Can't see exactly what you need?

We can perform custom differentiation runs from either our lines or your lines. Please ask us about our 'made-to-order' service.

axoCells™ Striatal Neurons

Striatal Neurons represent neurons from the human **striatum** which is related to movement control and reward. Striatal neurons progressively degenerate in patients with **Huntington's Disease** (HD).
They are frequently used for *in vitro* HD models. We supply them as NSCs with maturation to striatal neurons via our easy-to-follow protocol.

Assay ready in **31 days**

Express the **key markers** including DARPP32, CTIP2, CALBINDIN, and GABA

Designed for use in advanced ***in vitro*** **models**

Phenotypic characterization

We have extensively characterized our axoCells iPSC-derived striatal neurons using **immunocytochemistry** to identify the key striatal neuron markers (**fig. 1**).

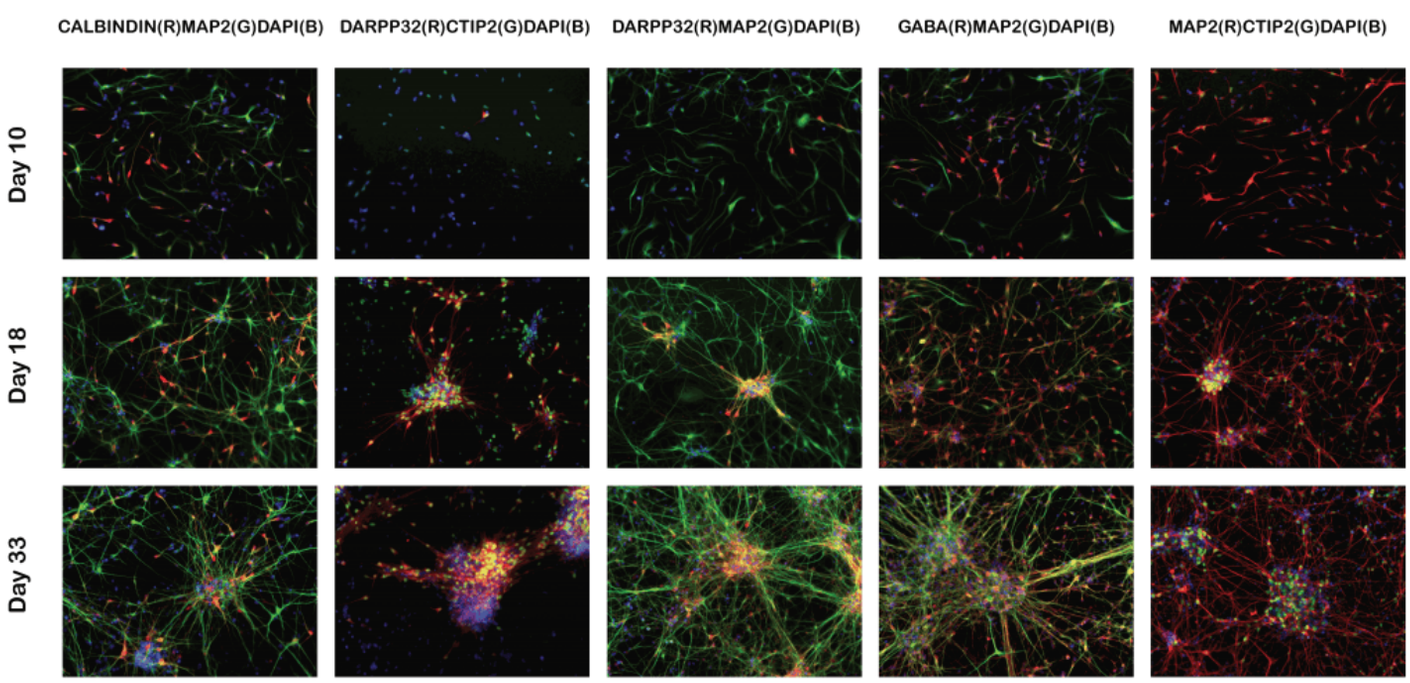


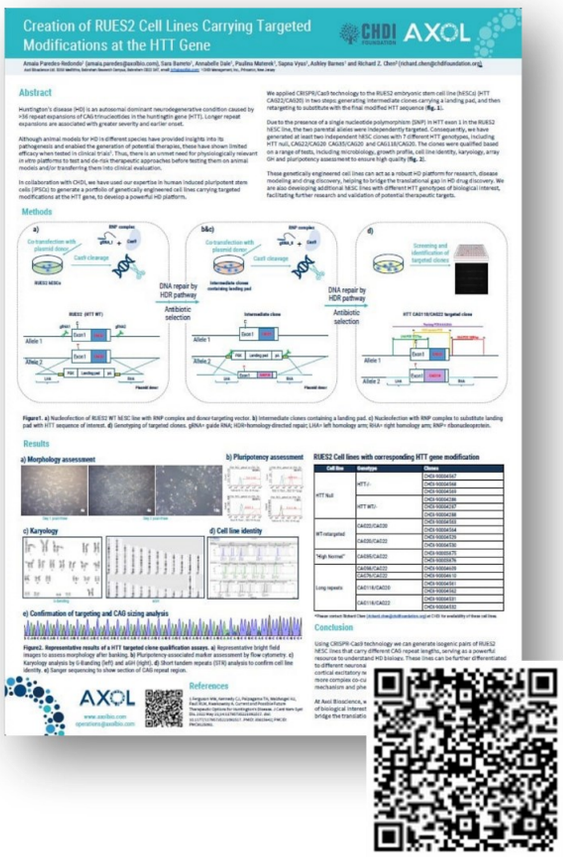
Figure 1. ICC images of differentiating axoCells Striatal Neurons. Cells were fixed and stained on day 10, day 18 and day 33 for key markers DARPP32, CTIP2, CALBINDIN, GABA and MAP2.

Creation of RUES2 Cell Lines Carrying Targeted Modifications at the HTT Gene

Huntington's disease (HD) is an autosomal dominant neurodegenerative condition caused by >36 repeat expansions of CAG trinucleotides in the huntingtin gene (HTT). Longer repeat expansions are associated with greater severity and earlier onset. Although animal models for HD in different species have provided insights into its pathogenesis and enabled the generation of potential therapies, these have shown **limited efficacy** when tested in clinical trials. Thus, there is an unmet need for **physiologically relevant *in vitro* platforms** to test and de-risk therapeutic approaches before testing them on animal models and/or transferring them into clinical evaluation.

In collaboration with CHDI, we have used our expertise in human iPSCs to generate a portfolio of genetically engineered cell lines carrying targeted modifications at the HTT gene, to develop a powerful HD platform.

Scan the QR code to access the poster.

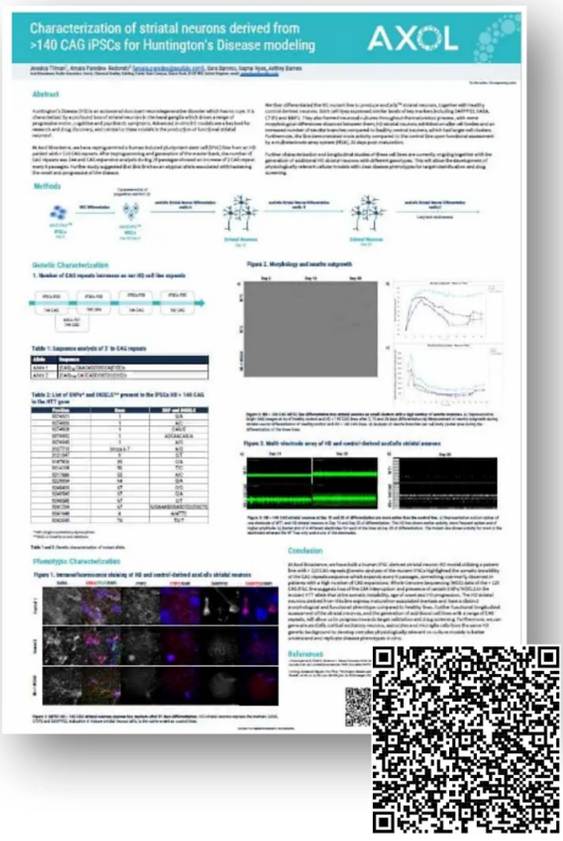


Characterization of striatal neurons derived from >140 CAG iPSCs for Huntington's Disease modeling

axoCells iPSC-derived striatal neurons are optimized for use in advanced *in vitro* models of **Huntington's Disease**. Central to this is our extensive experience with differentiating patient-derived iPSCs into high-quality striatal neurons.

In this poster, we describe the reprogramming of an iPSC line from an HD patient with **>120 CAG repeats**. After reprogramming and generation of the master bank, the number of CAG repeats was 144 and CAG expansion analysis during 15 passages showed an increase of 1 CAG repeat every 5 passages. Further study suggested that this line has an **atypical allele** associated with hastening the onset and progression of the disease.

Scan the QR code to access the poster.



axoCells Human iPSC-Derived Striatal Neurons

Supplied as Neural Stem Cells and a protocol to mature into final striatal neurons in **31 days**

Cells & Kits

Product Name	Cells only code / 1 vial	Quantity* / per vial	Kit** code
axoCells™ Human iPSC-Derived Neural Stem Cells, newborn male donor, ≥1.5 million cells	ax0015	≥1.5 million cells	**ax3115**
axoCells™ Human iPSC-Derived Neural Stem Cells, newborn female donor, ≥1.5 million cells	ax0016	≥1.5 million cells	**ax3116**
axoCells™ Human iPSC-Derived Neural Stem Cells, male donor, ≥1.5 million cells	ax0018	≥1.5 million cells	**ax3118**
axoCells™ Human iPSC-Derived Neural Stem Cells, Huntington's Disease (HTT CAG >50) female donor, ≥1.5 million cells	ax0211	≥1.5 million cells	**ax3211**

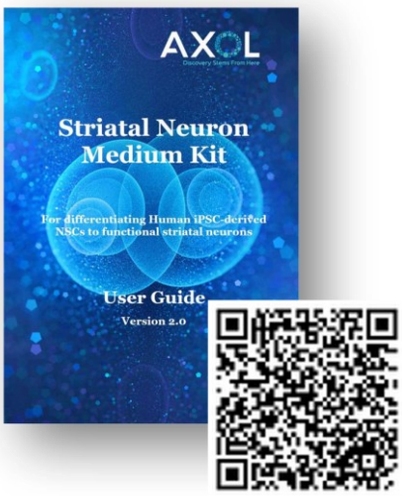
*Number of viable cells post thaw
**Kit contains cells and one of each item listed in the media and supplements table

Media and Supplements

Product Name	Product code	Quantity
axoCells™ Striatal Neuron Medium Kit, 500ml	ax0333	250 ml + 250 ml + 7.5 ml + 7.5 ml + 2 ml
axoCells™ Human BDNF Supplement, 10 µg	ax139800	10 µg
axoCells™ Human GDNF Supplement, 10 µg	ax139855	10 µg
axoCells™ SureBond-XF Coating, 1 ml	ax0053	1 ml

Additional third-party components may be required. Please refer to protocol for full list.

User Protocol



Can't see exactly what you need?

We can perform custom differentiation runs from either our lines or your lines. Please ask us about our 'made-to-order' service.

axoCells™ Cortical Inhibitory Interneurons

Cortical inhibitory interneurons are **GABAergic neurons** acting as the 'brakes' of the central nervous system. Connections between neurons in the brain are finely tuned and any increased electrical activity is dampened down by these cells. They are frequently used in **co-culture** methodologies for advanced *in vitro* models of **Alzheimer's Disease**.

Assay ready in **20** days

Express the **key markers** including GAD65, Parvalbumin-B, GABA and Somatostatin

Demonstrated **functionality** in advanced *in vitro* models including **co-culture and tri-culture**

Phenotypic characterization

We have extensively characterized our axoCells iPSC-derived cortical inhibitory interneurons using microscopy (**fig. 1**) and immunocytochemistry to identify the key markers (**fig.**2).

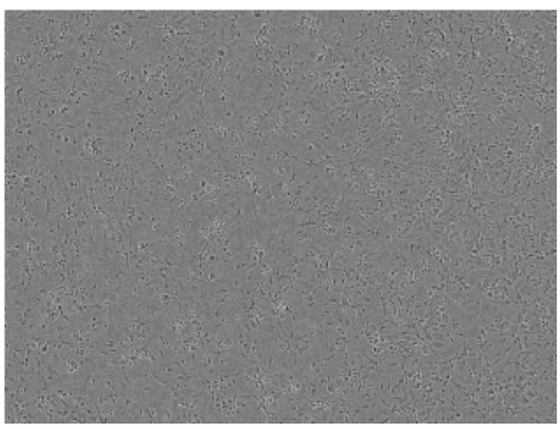


Figure 1. Phase contrast microscopy showing mature interneuron morphology at 150,000/cm^2 on day 20 in culture.

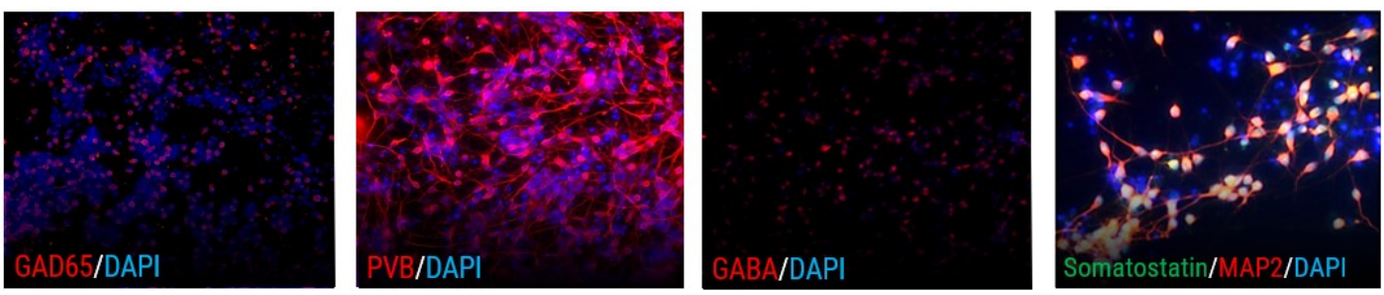


Figure 2. axoCells Cortical Inhibitory Interneurons (DIV20, ax0667) express the key markers GAD65, Parvalbumin (PVB), GABA, Somatastatin and MAP2.

axoCells Human iPSC-Derived Cortical Inhibitory Interneurons

Supplied as Neural Stem Cells and a protocol to mature into final striatal neurons in **20 days**

Cells

Product Name	Cells only code / 1 vial	Quantity* / per vial
axoCells™ Human iPSC-Derived Inhibitory Interneurons, new-born male donor, ≥2 million cells	ax0667	≥2 million cells
axoCells™ Human iPSC-Derived Inhibitory Interneurons, 40-50 yearr old male donor, ≥2 million cells	ax0662	≥2 million cells

*Number of viable cells post thaw
**Kit contains cells and one of each item listed in the media and supplements tab.

Media and Supplements

Product Name	Product code	Quantity
axoCells™ Neural Maintenance Media, 500ml	ax0031	500 ml
axoCells™ Human iPSC-Derived Cortical Neuron NeurOne Supplement	ax0674	1 ml + 1 ml
axoCells™ SureBond-XF Coating, 1 ml	ax0053	1 ml
axoCells™ Human BDNF Supplement, 10 µg	ax139800	10 µg
axoCells™ Human GDNF Supplement, 10µg	ax139855	10 µg
axoCells™ Neural Plating Media, 30ml	ax0033	30 ml

Additional third-party components may be required. Please refer to protocol for full list.

User Protocols

Can't see exactly what you need?

We can perform custom differentiation runs from either our lines or your lines. Please ask us about our 'made-to-order' service.

axoCells™ Microglia

Microglia are the **key immune cells of the brain** with crucial roles in brain development, neurogenesis, synaptic plasticity and homeostatic maintenance. They are frequently used in **co-culture** with neurons and muscle cells to **model AD and ALS**, and in monoculture for **compound screening.**

Assay ready in **7 days**

Express the **key markers** including IBA-1, TMEM119 and P2RY12

Demonstrate **robust functional activity**, measured by cytokine release, phagocytosis and chemotaxis

Neurodegenerative diseases affect around **15% of people worldwide**[1]**,** with **rising rates** due to our aging population. Research into these conditions has identified **neuroinflammation** as a **key disease driver**, so the drug discovery industry has turned to the **brain's main immune cell**, microglia, in search of new therapeutic targets.

First identified in the late 19th century by Frank Nissl (and his eponymous staining technique), microglia are becoming an **increasingly vital component** in human-relevant drug development platforms.

Microglia are the main immune cell of the central nervous system, making up around 10% of all cells in the brain[2]. They play key roles in brain development, neurogenesis, synaptic pruning and maintenance of the normal homeostatic environment[3]. Research has also revealed a key role for microglia in neurodegenerative diseases including **Alzheimer's Disease**, **Amyotrophic Lateral Sclerosis (ALS)** and **Parkinson's Disease**

Microglia derived from **human induced pluripotent stem cells (iPSCs)** can be produced **consistently** and incorporated into **model systems**. Human iPSC-derived microglia are made by differentiating iPSCs from reprogrammed donor blood samples. They therefore **retain the phenotypic characteristics** of the donor, producing a more **human-relevant** model system. This also opens up exciting applications of disease-derived microglia to test potential therapies in specific human disease models.

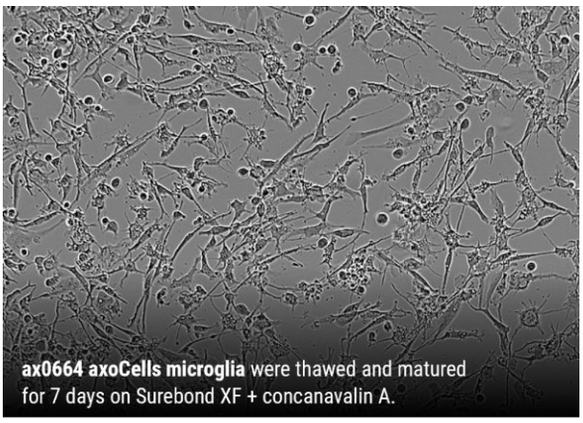
iPSC-derived microglia can also be produced in **large volumes**, optimizing cell consistency while adjusting the price point for larger-throughput platforms.

Advanced utility of axoCells iPSC-derived microglia

axoCells iPSC-derived microglia have **multiple applications** including *in vitro* models of Alzheimer's Disease and ALS. They can be used in a **variety of platforms**, from simple monoculture compound screens up to complex co-culture models, microfluidics systems or organ-on-chip models. As part of our **axoServices™** offering, we can develop microglia compound screening models to run in-house or tech transfer to customers.

1 Feigin V.L et al. doi: 10.1016/S1474-4422(19)30411-9.
2 Colonna M, Butovsky O. doi: 10.1146/annurev-immunol-051116-052358. Epub 2017 Feb 9. PMID: 28226226; PMCID: PMC8167938.
3 Gao, C et al. doi: 10.1038/s41392-023-01588-0

Phenotypic characterization



ax0664 axoCells microglia were thawed and matured for 7 days on Surebond XF + concanavalin A.

We've **extensively characterized** our axoCells Microglia to ensure the **correct morphology (fig. 1)** and expression of **key cell markers via immunocytochemistry (fig. 2) and flow cytometry (fig.3)**. We manufacture them to ISO 9001 standards to ensure **high quality and consistency**, at scale, to fuel robust *in vitro* neurodegenerative disease models.

Figure 1. axoCells Microglia showing expected morphology, with different phenotypes demonstrating sub-populations with specified functions.

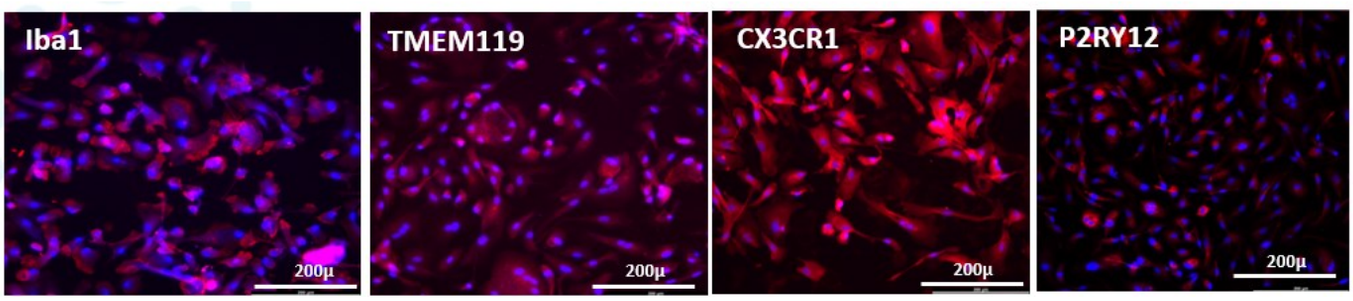


Figure 2. Immunocytochemistry of axoCells Microglia demonstrating the expression of key markers Iba1, TMEM119, CX3CR1 and P2RY12.

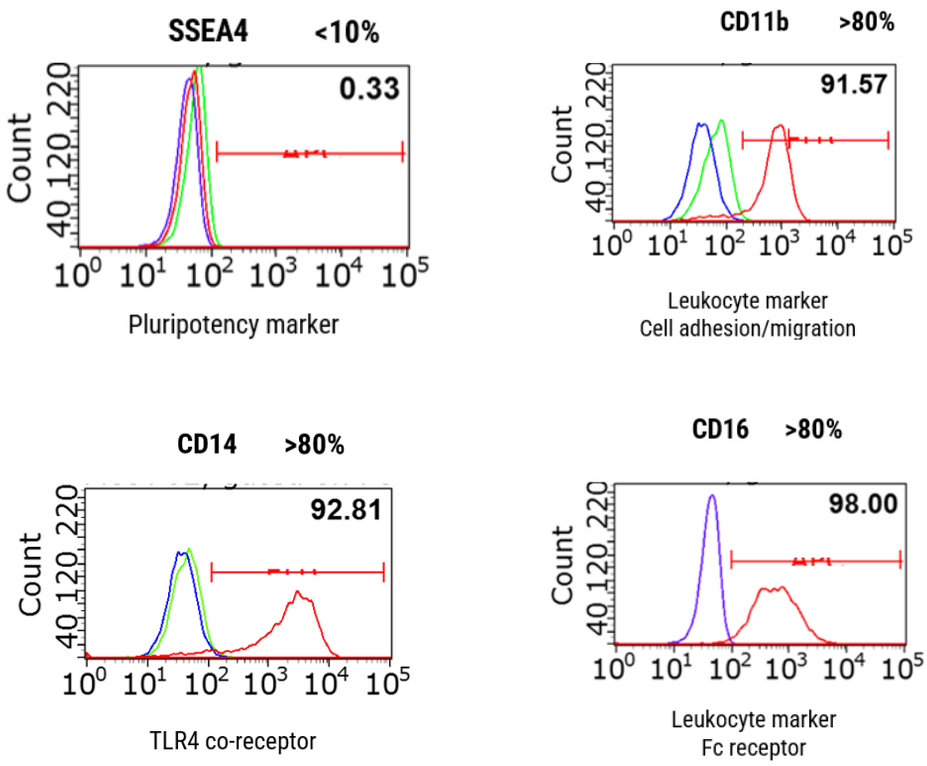
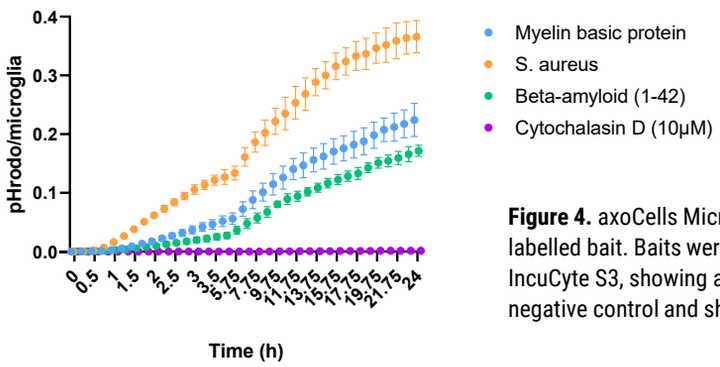


Figure.3 Example flow cytometry QC for fresh macrophage progenitors, demonstrating the presence of lineage-specific markers CD14, CD11b and CD16 above threshold levels, and negative control SSEA4 below threshold. This batch would therefore pass this stage of QC. Our standard panel also includes CD206 and CD163 as lineage-specific markers. Unstained, Isotype (control), Marker of interest.

Functional characterization

We've performed a range of assays to assess the **functional performance** of our axoCells Microglia, including phagocytosis (fig.4), chemotaxis (fig. 5) and cytokine release (fig. 6).

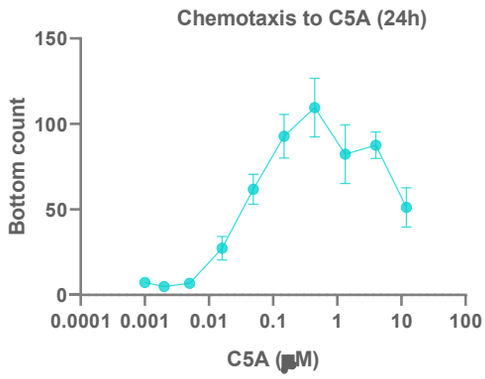
Phagocytosis



axoCells Microglia exhibit phagocytosis of various baits including myelin basic protein, *S. aureus* and beta-amyloid. This activity is inhibited by cytochalasin D.

Figure 4. axoCells Microglia were thawed and matured for 7 days before addition of pHrodo labelled bait. Baits were added to the cells and phagocytosis monitored over 24h using an IncuCyte S3, showing a steady increase over time. Cytochalasin D (10uM) was used as a negative control and showed complete inhibition of phagocytosis.

Chemotaxis



axoCells Microglia demonstrate expected chemotaxis to various concentrations of C5a, with very large concentrations exhibiting an expected inhibitory effect.

Figure 5. Chemotaxis by fresh axoCells Microglia to various concentrations of C5a. iPSC-derived microglia were matured for 7 days before re-plating into chemotaxis plates. Cell movement was measured using an IncuCyte S3. Values represent the number of cells moving from the top chamber to the bottom, towards C5a, after 24h. Data are n=4 +/- SEM.

Cytokine release

axoCells Microglia exhibit the expected pattern of functional cytokine release.

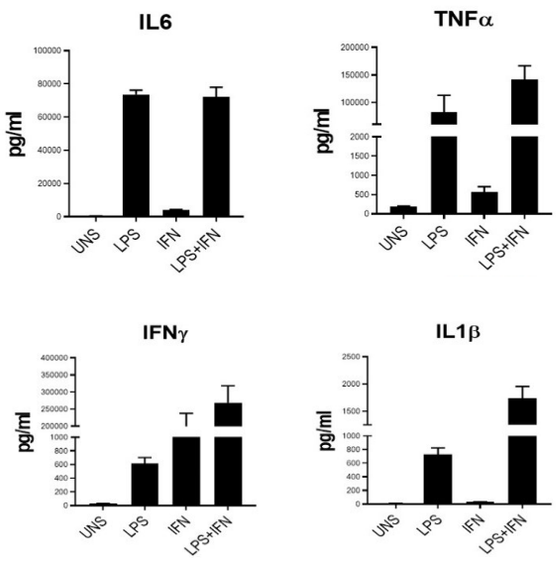


Figure 6. Cytokine release from fresh axoCells Microglia following 24 h stimulation with LPS, INFy or both (n=3). UNS= unstimulated. The expected pattern of cytokine release demonstrates functional relevance.

Fuel high performance in assay systems

axoCells Microglia have been used extensively to fuel advanced *in vitro* assay systems. They have also featured in **over 40% of our custom axoServices project work**, used in monoculture or in co-culture with other neuronal cells for research and compound screening projects.

If you would like to power your assay system with our functional iPSC-derived microglia, please get in contact at operations@axolbio.com

How we bulk manufacture high quality microglia

From our manufacturing facility in Roslin, we work to ISO 9001 standards to ensure our cells are **high-performing**, **functionally relevant** and **consistent** - even in bulk quantities. We utilize **rigorous QC** across the following parameters:

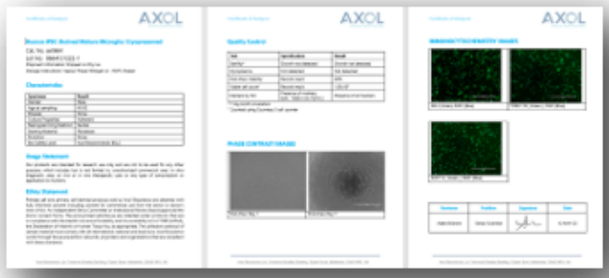
Test	Specification
Flow Cytometry	Presence of lineage-specific markers and absence of pluripotency marker
Sterility*	Growth not detected
Mycoplasma	Not detected
Post-thaw Viability	Record result
Viable Cell Count**	Record result
Markers by ICC	Presence of markers: IBA1, TMEM119, P2RY12, CX3CR1

*7 day broth incubation
**Counted using Countess™ 3 Automated Cell Counter

We use flow cytometry to assess progenitor cells for a standard panel of lineage-specific markers, ensuring **high levels of quality control**. SSEA4 is used as a negative control to detect markers of pluripotency.

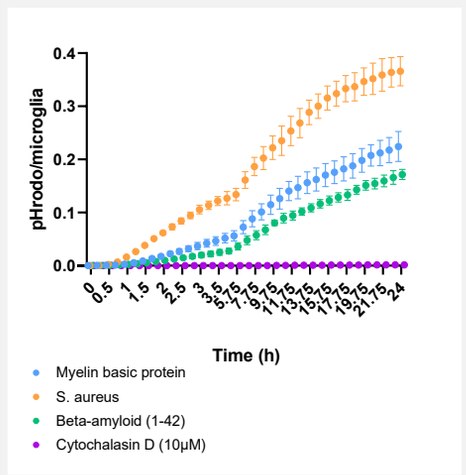
Supporting quality and consistency

We want our customers to have confidence in using our microglia in their neural workflows. That's why our production facility works to **ISO 9001 standards**, guided by our rigorous quality control procedures and decades of scientific experience. All cells come with a full **Certificate of Analysis** (shown right) and are officially certified by HPSCreg®, to ensure ethical and biological conformity for your peace of mind.



Functional QC for axoCells Microglia

We've been looking into **functional QC (fQC)** as the next step in the quality chain, testing the **utility and performance** of cells in biologically-relevant assays. We believe this new standard will enhance **confidence** in the physiological relevance of our cells and drive **better translational power** in advanced *in vitro* models.



axoCells Microglia can power advanced *in vitro* models for Alzheimer's Disease and other neurodegenerative diseases. fQC will include a phagocytosis assay (fig. 7) measuring bait uptake over 24 hours against a pre-determined threshold, with inhibition when adding cytochalasin D.

Figure 7. axoCells Microglia demonstrating phagocytosis of various baits. axoCells Microglia were thawed and matured for 7 days before addition of pHrodo labelled bait. Baits were added to the cells and phagocytosis monitored over 24h using an IncuCyte® S3, showing a steady increase over time. Cytochalasin D (10µM) was used as a negative control and showed complete inhibition of phagocytosis.

The utility of axoCells Microglia

We manufacture functional, consistent microglia for multiple applications including **monoculture compound screening** and in **co-culture** with neuronal and muscle cells for **ALS and Alzheimer's Disease models.**

Phagocytosis assays

A key functional role of microglia is the ability to perform phagocytosis in the brain. We have **extensively characterized** our phagocytosis assays with a **wide range** of pHrodo-labelled baits, including *E. coli*, zymosan beads, beta-amyloid, TAU, myelin basic protein, dead neurons, alpha-synuclein and *S. aureus*. This enables us to **select appropriate baits based on experimental need**, including disease-associated baits such as **alpha-synuclein (important for Parkinson's Disease modelling) and beta-amyloid (relevant for Alzheimer's Disease). Fig. 8** demonstrates a phagocytosis assay comparing health control-derived microglia with ALS-derived (C9ORF72) microglia.

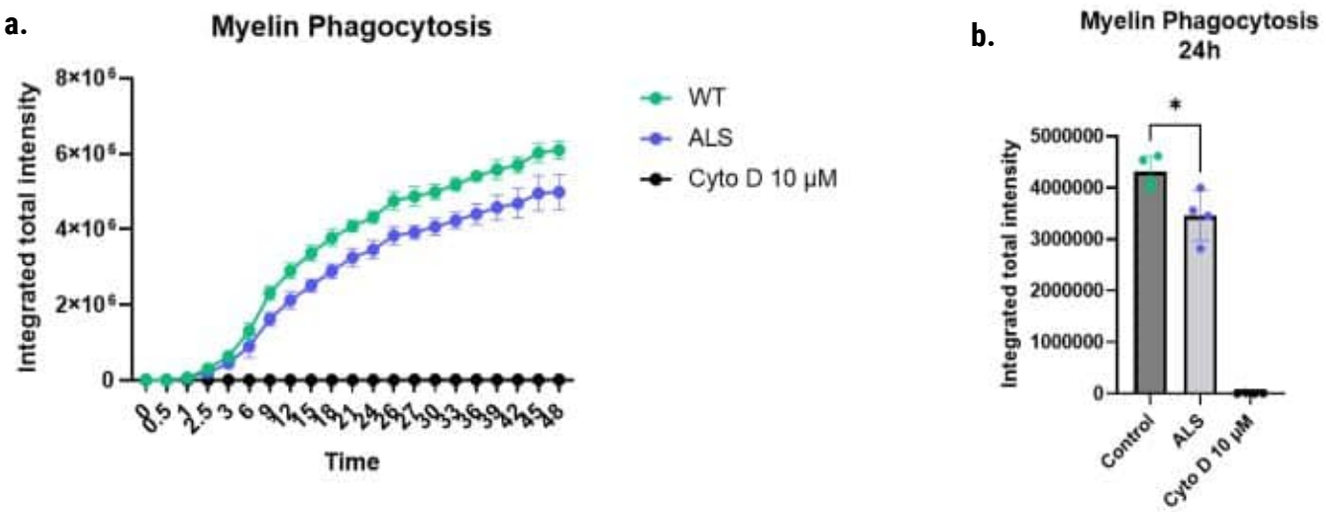


Figure 8. Fresh iPSC-derived microglia from healthy or ALS (C9orf72) background were matured for 7 days and phagocytosis of myelin basic protein (MBP) assessed. MBP was labelled with pHrodo dye and added to the cells. **7a.** Phagocytosis was quantified using an IncuCyte S3 for up to 48h . **7b.** Data after 24h from addition of bait. A t-test was perfomed to assess any statistical significance between the cell lines * $p<0.05$.

Compound screening assays

A crucial component of any compound screening project is generating **high-quality assay data**. With microglia featuring in over 40% of our axoServices projects, we've developed the following assays for microglia compound screening:

Real-time imaging	Electrophysiology (multi-electrode array)
• Phagocytosis • Chemotaxis • Cytokine release	Measurement of neuronal activity in co-culture models

Case studies

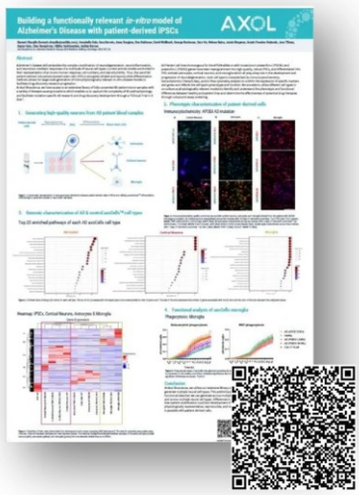
axoCells Microglia have been used extensively in a **range of model systems**. With rapid maturation times, robust functional performance and guided by over a decade of iPSC expertise, we enjoy collaborating with groups looking to unlock the benefits of iPSC-derived microglia models.

Powering high content imaging workflows with Sygnature Discovery

We recently announced our collaboration with Sygnature Discovery to incorporate **axoCells human iPSC-derived microglia** into their high-content imaging *in vitro* screening workflows. This will enable researchers to gain a **deeper understanding** of the cellular response to drug candidates and identify **potential therapeutic targets** with higher precision and accuracy.

> *Our collaboration highlights the importance of leveraging **innovative technologies** and **expertise** to enhance the efficiency and effectiveness of the drug discovery process. Axol's extensive practical experience in handling human iPSCs will **accelerate our ambition** to offer high-content-based imaging assays to customers in the neuroscience therapeutic area.*
>
> - **Tim Phillips,** Associate Director (Bioscience) at Sygnature Discovery

Building a functionally relevant *in vitro* model of Alzheimer's Disease with patient-derived iPSCs



Here we describe the characterization of **astrocytes**, **cortical excitatory neurons** and **microglia** derived from Alzheimer's Disease patient cell lines homozygous for the APOE4 allele or with mutations in presenilin-1 (PSEN1) and presenilin-2 (PSEN2) genes.

These cell types can be used in co-culture to build advanced *in vitro* models of neurodegeneration for research and drug discovery.

Scan the QR code to download the poster.

Assessing the functional performance of ALS-derived microglia

In this exciting white paper in collaboration with Sartorius, we compared the **morphological and functional performance** of healthy control-derived and ALS-derived axoCells Motor Neurons and microglia.

In this project, we demonstrated **distinct ALS-like phenotypes** in axoCells Motor Neurons and microglia derived from iPSCs generated from ALS patient donor cells, which can be quantified and used to inform future drug discovery purposes. We also looked into **morphology**, **immunocytochemistry** and **electrophysiology** (data available in the full whitepaper), demonstrating the wide range of useful assays for investigating ALS-like phenotypes in motor neurons and microglia.

Scan the QR code to read the full whitepaper.

axoCells Human iPSC-Derived Microglia

Supplied as Neural Stem Cells and a protocol to mature into final striatal neurons in **7 days**

Cells & Kits

Product Name	Cells only code / 1 vial	Quantity* / per vial	Kit** code
axoCells™ Human iPSC-Derived Microglia, male donor, ≥1 million cells	ax0664	≥1 million cells	**ax0679**

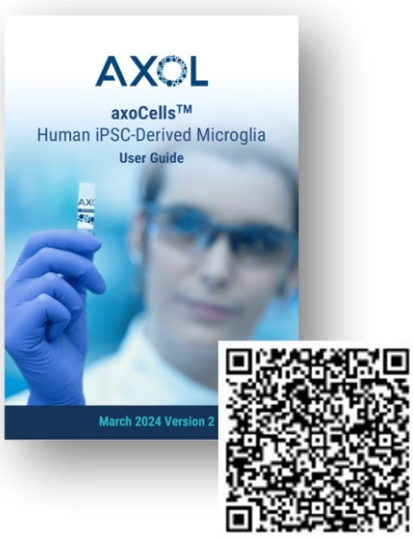
*Number of viable cells post thaw
**Kit contains cells and one of each item listed in the media and supplements table

Media and Supplements

Product Name	Product code	Quantity
axoCells™ Human iPSC-Derived Microglia Media and Supplement kit	ax0660	100 mL + 100 µL + 100 µL + 1 mL
axoCells™ SureBond-XF Coating, 1 ml	ax0053	1 ml

Additional third-party components may be required. Please refer to protocol for full list.

User Protocol



Can't see exactly what you need?

We can perform custom differentiation runs from either our lines or your lines. Please ask us about our 'made-to-order' service.

axoCells™ Astrocytes

axoCells iPSC-derived astrocytes are a subtype of glial (supportive) cell and play **critical roles** in synapse function, synaptic remodeling and the regulation of blood flow. These cells are **optimized for co-culture** with neurons and neuroinflammatory cells for advanced *in vitro* models of Alzheimer's Disease and ALS.

Assay ready in **2 days**

Express the **key markers** (GFAP and S100) with low expression of Nestin (a NSC marker) and TUJ1 (a neuronal marker)

Designed for use in advanced ***in vitro*** **models**

Phenotypic characterization

We have extensively characterized our axoCells iPSC-derived astrocytes using immunocytochemistry to identify the key astrocyte markers (**fig. 1**).

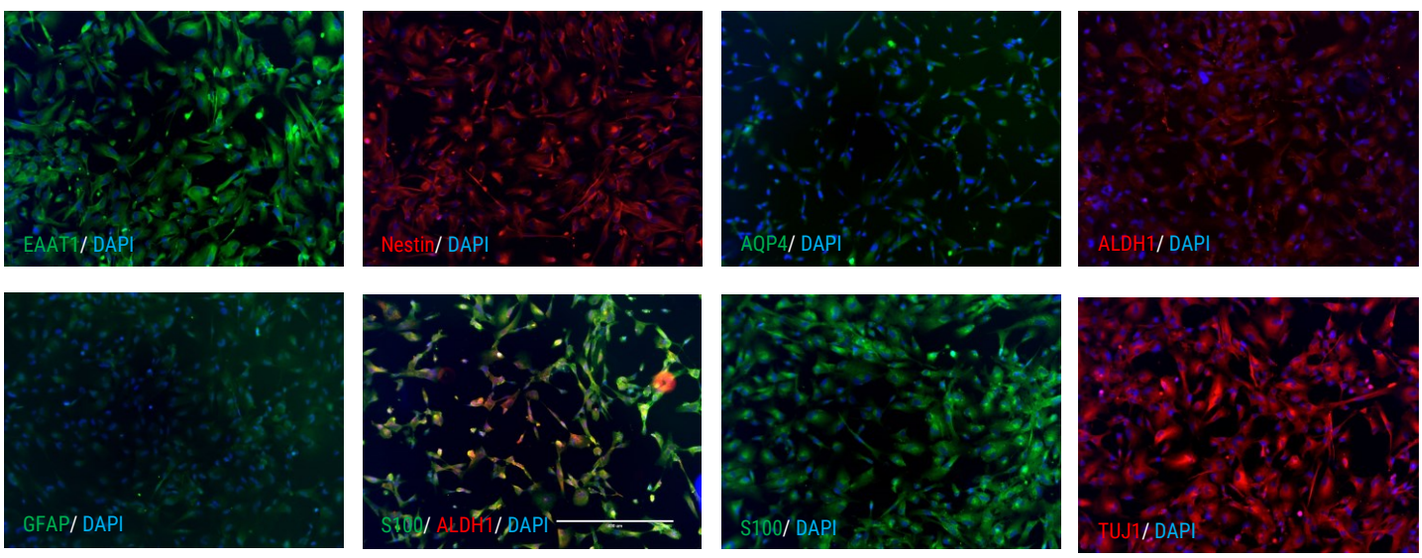


Figure 1. axoCells iPSC-derived astrocytes (ax0704) express key astrocyte-specific markers (GFAP, AQP4 and S100B) and astrocyte-associated markers, EAAT1 and ALDH1L1, while showing very low levels of neuronal progenitor markers such as Nestin.

axoCells Human iPSC-Derived Astrocytes

Supplied as Neural Stem Cells and a protocol to mature into final striatal neurons in **2 days**

Cells & Kits

Product Name	Cells only code / 1 vial	Quantity* / per vial
axoCells™ Human iPSC-Derived Astrocytes, male donor, ≥1 million cells	ax0704	≥1 million cells

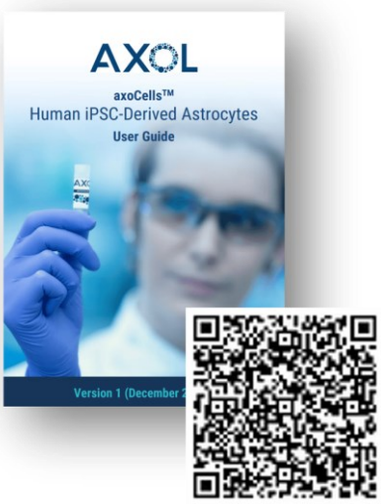
*Number of viable cells post thaw

Media and Supplements

Product Name	Product code	Quantity
axoCells™ SureBond-XF Coating, 1 ml	ax0053	1 ml
axoCells™ Human FGF2, 100 µg	ax0047	100 µg

Additional third-party components may be required. Please refer to protocol for full list.

User Protocols



Can't see exactly what you need?

We can perform custom differentiation runs from either our lines or your lines. Please ask us about our 'made-to-order' service.

axoCells™ Myotubes

axoCells human iPSC-derived myotubes can be matured into **skeletal muscle cells** for use in **advanced *in vitro* musculoskeletal and neuromuscular model systems**. They are frequently used in **advanced *in vitro* models of the neuromuscular junction** and for **ALS disease modeling**.

Assay ready in **5 days***

*May vary if patient disease lines are used

Expression of **key proteins** including Desmin, Dystrophin, Myosin Heavy Chain and Titin

Demonstrated **functionality** in advanced *in vitro* models

axoCells myotubes - thaw to assay-ready in just 5 days

axoCells iPSC-derived myotubes are supplied as frozen myogenic progenitors with a rapid 5-day thaw-to-assay protocol.

axoCells myotubes mature to form elongated, striated, multinucleated cells expressing the key proteins Desmin, Dystrophin, Titin, and Myosin Heavy Chain (**fig.1**). They have been specifically developed for use in advanced *in vitro* musculoskeletal and neuromuscular model systems.

We supply you with frozen myogenic progenitors that mature into fused myotubes in just 5 days

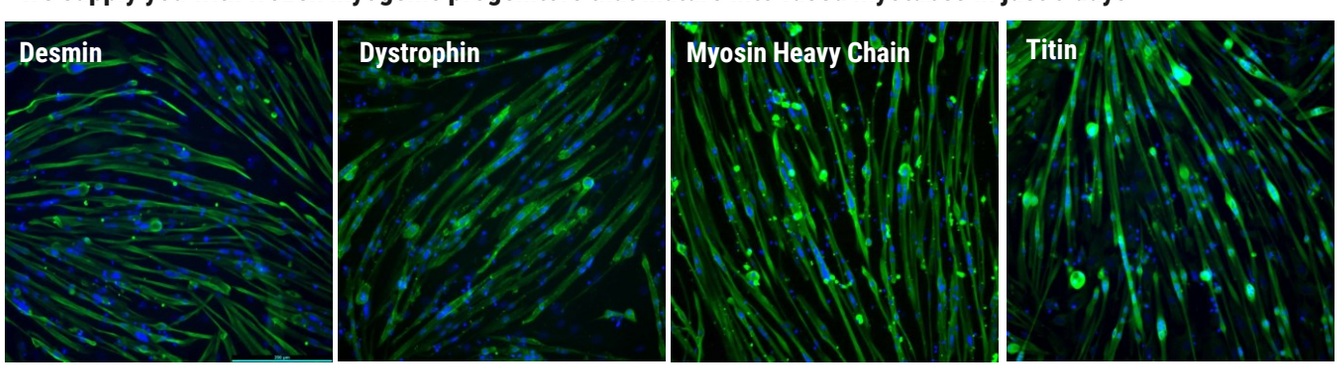


Figure 1. ax3062 fused myotubes stained after 5 days for DAPI (blue) and Desmin, Dystrophin, Myosin Heavy Chain and Titin (all green). Images at 20x magnification.

How to get high quality axoCells™ myotubes

Myotubes are available “made–to–order“ with a 10–week turnaround. Note, that duration may vary depending if patient disease lines are used. Standard delivery is 10 vials per run (but can be specified at time of ordering)

Applications

axoCells Myotubes have been specifically developed for use in advanced *in vitro* musculoskeletal and neuromuscular model systems, including the neuromuscular junction and ALS.

Available as a custom “made-to-order” production run only. Contact operations@axolbio.com for further information.

axoCells™ Motor Neurons

Motor neurons innervate muscle cells to control a range of voluntary and involuntary movements. The progressive destruction of motor neurons is central to neuromuscular conditions including **ALS**. Our axoCells Motor Neurons are frequently used together with muscle cells (axoCells Myotubes) to fuel **advanced *in vitro* models of ALS**.

Assay ready in **10 days**

Express the **key markers** including HB9, MAP2, LIM3 and ChAT2

Demonstrated **functionality** in advanced *in vitro* models

Phenotypic characterization

We've **extensively characterized** our axoCells Motor Neurons for phenotypic relevance including **correct morphology (fig. 1)** and expression of **key cell markers via immunocytochemistry (fig. 2).**

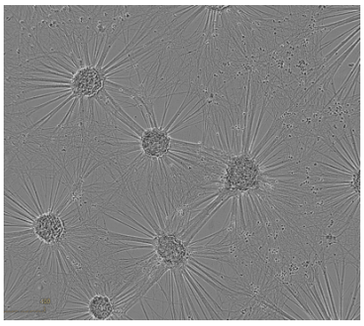


Figure 1. Phase contrast images of axoCells Motor Neurons matured from progenitors over 21 days.

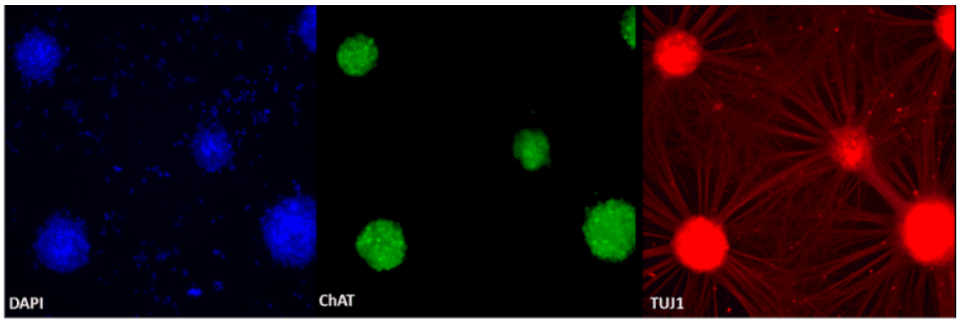
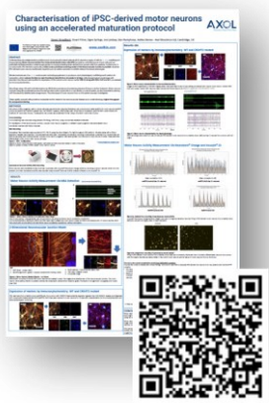


Figure 2. Immunocytochemistry of day 21 mature axoCells Motor Neurons demonstrating presence of key markers (ChAT and TUJ1). Images captured on a Leica microscope x20 magnification.

Enhanced maturation with our Accelerator Supplement

We've developed an *in vivo* environment-mimicking supplement to reduce maturation times for our axoCells Motor Neurons from six weeks to just 10 days, with phenotypic and functional activity as assessed by morphology, immunocytochemistry and electrophysiology.

Scan the QR code to explore the data behind the development of our Accelerator Supplement.



Functional relevance

We've validated the **functional relevance** of our axoCells Motor Neurons across a range of assays including **electrophysiology** and **calcium imaging (fig.3) with disease phenotypes (fig.4)**.

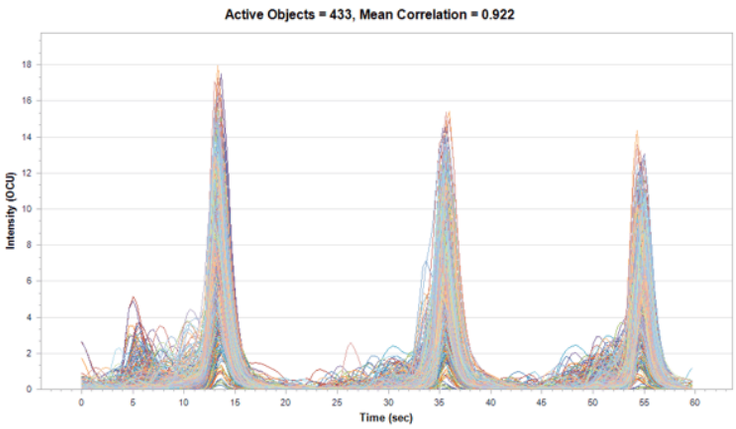


Figure 3. Firing pattern of axoCells Motor Neurons at day 21, transfected with NeuroBurst (calcium-sensitive lentivirus driven off the synapsin reporter) to pick up spontaneous neuronal activity on an IncuCyte. This demonstrates regular, synchronized firing with a mean correlation of 0.92. Potential parameters measured include mean correlation, burst rate, burst duration and burst strength.

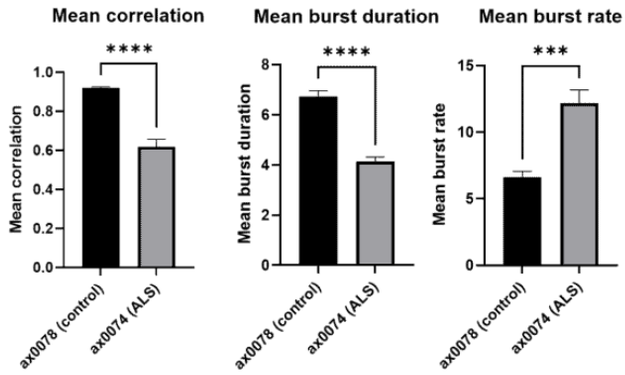
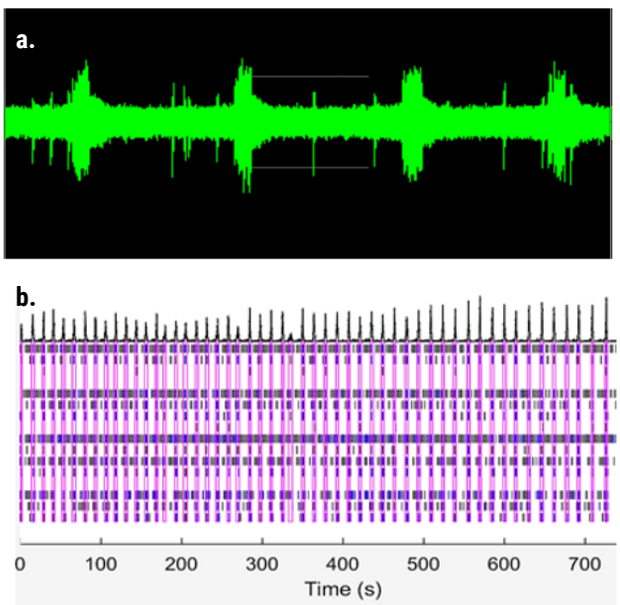


Figure 4. axoCells healthy control and ALS-derived motor neurons (C9ORF72) were matured for 21 days and transfected with Neuroburst Orange to look at spontaneous neuronal activity on the IncuCyte S3. Healthy motor neurons show regular synchronous firing compared to ALS which fire more frequently, for shorter duration and in a less-synchronized manner (shown by burst duration and rate). N=3, ***p<0.001, ****p<0.0001

Functional QC

We've been looking into **functional QC (fQC)** as the next step in the quality chain, testing the **utility and performance** of cells in biologically-relevant assays. We believe this new standard will enhance **confidence** in the physiological relevance of our cells and drive **better translational power** in advanced *in vitro* models.



Advanced *in vitro* neuromuscular models, fueled by axoCells Motor Neurons, can be used for ALS drug discovery. fQC will use assays that measure the appearance of synchronized burst firing via multi-electrode array (fig. 5), which would represent functional network formation.

Figure 5. axoCells Motor Neurons demonstrating synchronized burst firing at day 10, measured on the Axion Maestro Pro MEA system. Sodium spike firing and network burst firing responses of the motor neurons were observed. **5a.** Sodium spike profile; **5b.** Raster plot showing burst firing events (blue boxes) with synchronized firing highlighted in pink boxes.

axoCells Human iPSC-Derived Motor Neurons

Supplied as Neural Stem Cells and a protocol to mature into final striatal neurons in **10 days**

Cells & Kits

Product Name	Cells only code / 1 vial	Quantity* / per vial	Kit** code
axoCells™ Human iPSC-Derived Motor Neurons, male donor, ≥2 million cells	ax0078	≥2 million cells	**ax0178**
axoCells™ Human iPSC-Derived Motor Neurons, ALS (C9ORF72) asymptomatic male donor, ≥2 million cells	ax0073	≥2 million cells	-
axoCells™ Human iPSC-Derived Motor Neurons, ALS (C9ORF72) female donor, ≥2 million cells	ax0074	≥2 million cells	-

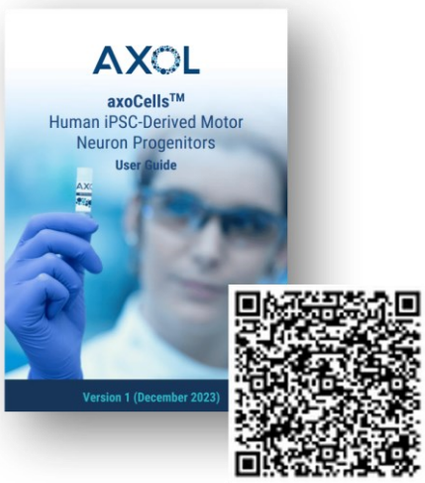
*Number of viable cells post thaw
**Kit contains cells and one of each item listed in the media and supplements table

Media and Supplements

Product Name	Product code	Quantity
axoCells™ Human CNTF Supplement, 20 µg	ax139888	20 µg
axoCells™ Human GDNF Supplement, 10µg	ax139855	10 µg
axoCells™ Human BDNF Supplement, 10 µg	ax139800	10 µg
axoCells™ Motor Neuron Maintenance Media, 200 ml	ax0072	200 ml
axoCells™ Motor Neuron Accelerator Supplement, 1 ml	ax0179	1 ml

Additional third-party components may be required. Please refer to protocol for full list.

User Protocol



Can't see exactly what you need?

We can perform custom differentiation runs from either our lines or your lines. Please ask us about our 'made-to-order' service.

Pain & Sensation

Building better *in vitro* pain, sensory and peripheral neurotoxicity models with human iPSC technology

Pain & Sensation

iPSC models for pain and peripheral neurotoxicity studies

Sensory neurons

Chronic pain disorders have been estimated to affect **over 30% of people worldwide**, causing significant physical, emotional and economic burden[1]. Alongside this, drug discovery has encountered significant regulatory clearance challenges from **adverse neurotoxic effects**, particularly in **chemotherapy-induced peripheral neuropathy**[2].

This has driven researchers to develop various model systems including **animal-based platforms** and **simple cell cultures** comprising rodent dorsal root ganglion neurons[2]. While these models have provided some valuable insights, there remains a **translational gap** from bench to bedside that has hindered drug discovery and toxicology. The industry has therefore turned to more **human-relevant** model systems powered by **human iPSC-derived cells.**

1 Cohen et al. 2021 doi: 10.1016/S0140-6736(21)00393-7
2 Xiong, C. et al. 2021 doi: 10.1111/cts.12912

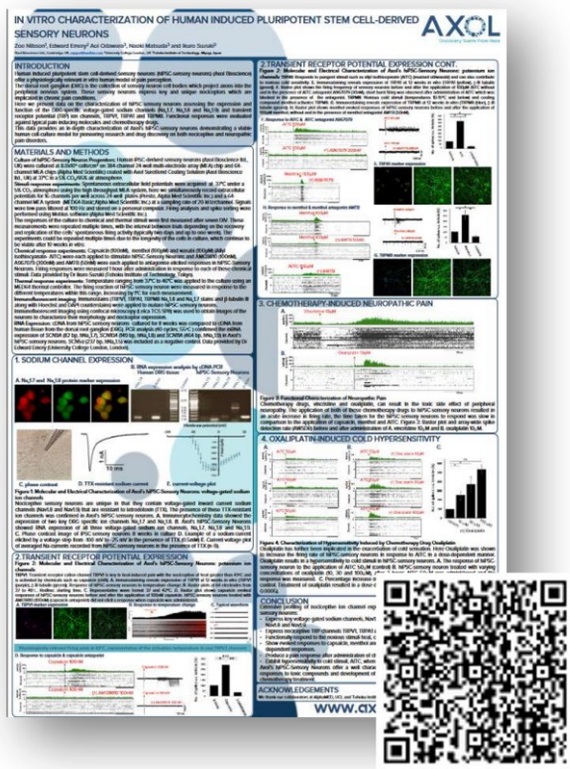
Scan the QR code to read our poster **"*In vitro* characterization of human induced pluripotent stem cell-derived sensory neurons"** including data on key ion channel expression, capsaicin challenge and chemotherapy-induced neuropathic pain models.

We have specifically developed a range of **axoCells iPSC-derived sensory neurons** for use in neurodegenerative disease models.

Key highlights include:

- Expression of the **key nociceptive ion channels** including $Na_v1.7$ and the DRG-specific, TTX-resistant channels, $Na_v1.8$ and $Na_v1.9$ as well as the temperature-sensitive, TRPV1 and TRPM8, and TRPA1, a sensor of pungency, bitterness and cold
- **Functional relevance** across multiple assays including capsaicin and menthol treatment, thermoception and neurite outgrowth with paclitaxel
- Designed for **advanced *in vitro* model formats** including co-culture, microfluidics devices and organ-on-chip platforms
- Manufactured in our **ISO 9001-accredited** production facility with excellent **ISSCR Standards compliance**

With our axoCells iPSC-derived sensory neurons, you can unlock the benefits of iPSC technology for your *in vitro* pain and peripheral neurotoxicity models.

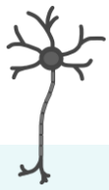


axoCells™ Sensory Neurons

Sensory neurons are the nerve cells activated by sensory input from the environment, including **touch, heat and pain**. They are frequently used in **cosmetic testing, pain models and peripheral neurotoxicity models**, often in microfluidic devices.

Assay ready in **21 days**

Express 50 of the **key ion channels** including Nav1.7, Nav1.8, Nav1.9, TRPV1 and TRPA1



Designed for use in **microfluidics platforms** and **organ-on-chip devices** for models of pain and peripheral neurotoxicity

Phenotypic characterization

We have extensively characterized our axoCells iPSC-derived sensory neurons using **immunocytochemistry** to identify the key markers (**fig.1**).

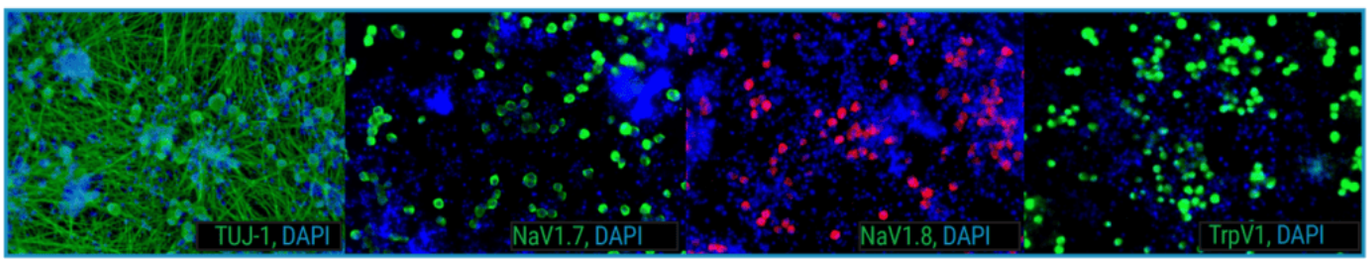


Figure 1. Immunocytochemistry of axoCells Sensory Neurons show expression of the key marker TUJ-1, indicative of neuronal development, and the nociceptive ion channels TrpV1, $Na_v1.7$ and $Na_v1.8$, responsible for the generation and maintenance of abnormal neuronal electrogenesis and hyperexcitability in the development of pathological pain.

Functional relevance

We have performed extensive validation of our axoCells iPSC-derived sensory neurons across numerous assays including capsaicin and menthol challenge (**fig. 2**), thermoception (**fig.3**) and neurite outgrowth (**fig. 4**).

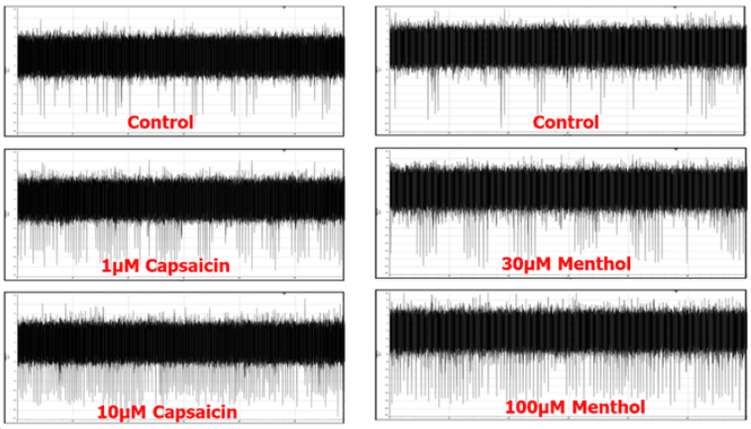


Figure 2. axoCells Sensory Neurons were challenged with capsaicin and menthol at 22 and 27 days respectively. As early as 22 days, 90% of the neurons responded to capsaicin challenge with increased spike measurements on a multi-electrode array (MEA) platform. Dose responses for both chemicals were observed, demonstrating their value in advanced *in vitro* models of pain, sensation, and peripheral neurotoxicity.

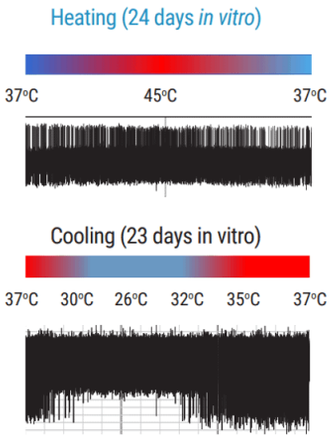


Figure 3. Electrophysiology recordings demonstrating increased firing of axoCells Sensory Neurons in response to heating and decreased firing in response to cooling. This demonstrates thermoception, an *in vivo* function, and shows the functional relevance of axoCells Sensory Neurons.

Treatment of axoCells Sensory Neurons with the chemotherapy reagent **paclitaxel** results in **reduced neurite length** (axotomy) demonstrating the functional relevance of the sensory neurons (**fig.4**). This provides a model for both **acute insult** and **chronic peripheral neurotoxicity**.

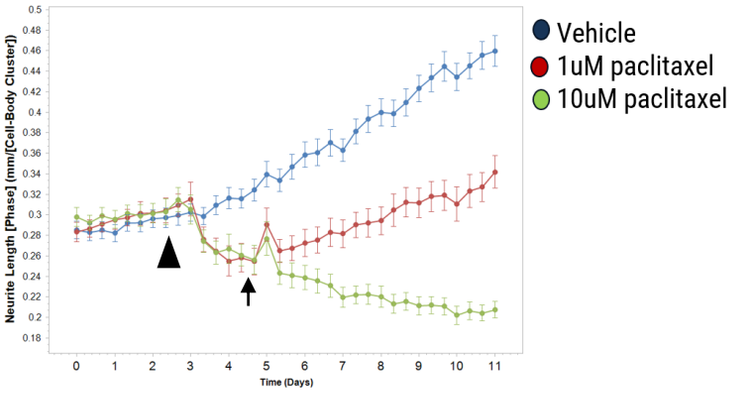


Figure 4. Response of day 25 axoCells Sensory Neurons to paclitaxel, a chemotherapy reagent, applied at 2.6 days (arrowhead) with washoff at 4.6 days (arrow)

fQC for axoCells Sensory Neurons

We've been looking into **functional QC (fQC)** as the next step in the quality chain, testing the **utility and performance** of cells in biologically-relevant assays. We believe this new standard will enhance **confidence** in the physiological relevance of our cells and drive **better translational power** in advanced *in vitro* models.

axoCells Sensory Neurons are optimized for use in *in vitro* pain models. fQC will measure the response to capsaicin at 1µM and 10µM concentrations via multi-electrode array.

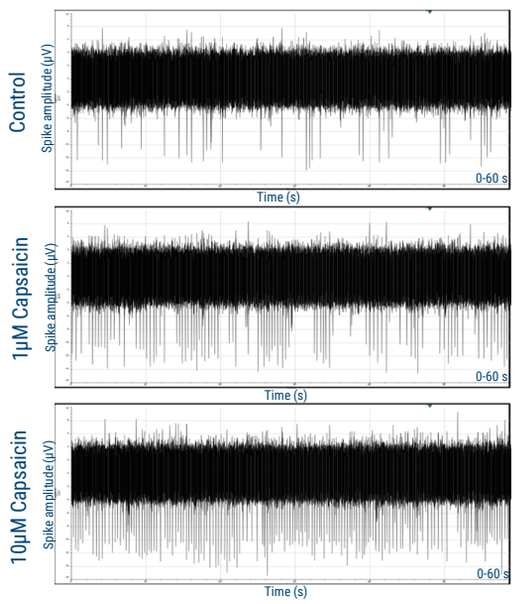


Figure 5. axoCells Sensory Neurons demonstrating response to 1µM and 10µM capsaicin at 22 days, measured on the Axion Maestro Pro multi-electrode array (MEA) system.

Accelerated maturation with our Maximizer Supplement

Our Maturation Maximizer media supplement ensures **faster maturation** of our iPSC-derived sensory neurons compared to those cultured in our traditional maintenance media. It works by **mimicking *in vivo* signals** between sensory neurons and their supporting cells. The supplement contains signaling factors present in the peripheral nervous system and, in particular, the native environment of sensory neurons.

Utilizing this supplement **accelerates the maturation** of iPSC-derived sensory neurons to drive **mature, functional neurons in just 21 days**. It also raises the **basal electrical activity** which is useful for assays measuring inhibition of sensory neuron activity (**fig. 6**).

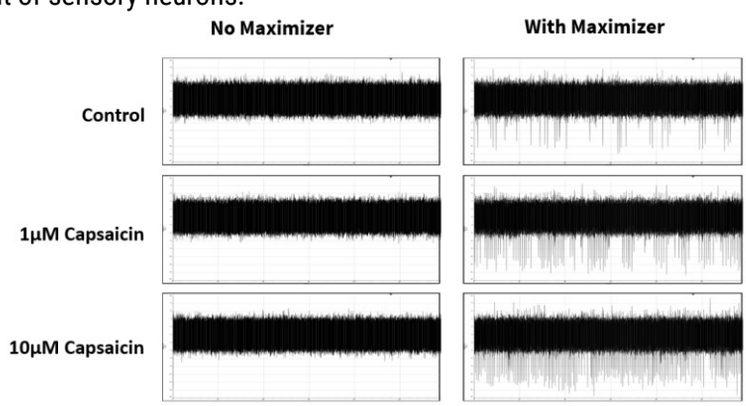


Figure 6. Electrophysiology responses comparing axoCells Sensory Neurons grown with and without Maximizer supplement, with increasing concentrations of capsaicin, measured at day 22 in culture.

Case study:
Powering microfluidics devices with axoCells Sensory Neurons

axoCells Sensory Neurons are being used as the "fuel" to power NETRI's high-throughput NeuroFluidics devices for pre-clinical neuroscience research, cosmetics testing and drug discovery. These organs-on-chip (OOC) kits facilitate the use of dedicated disease models for conditions including **pain** and **nerve injury**. By incorporating high-quality axoCells Sensory Neurons into NETRI's compartmentalized and MEA-compatible OOC devices, these kits can offer researchers **better predictivity** and **translational outcomes**.

At Axol, we're excited to see the launch of this organ-on-chip (OOC) range, which will be powered by axoCells iPSC-derived sensory neurons. Combining the **quality and consistency** of Axol's sensory neurons with NETRI's OOC capabilities, the company will be able to deliver **robust *in vitro* model systems** of the peripheral nervous system.

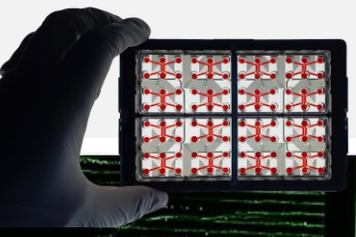
Built in a **high-throughput format**, these OOC kits will enable the generation of **larger, faster, and more predictive datasets**. Ultimately, this collaboration will help to drive the adoption of OOCs in biopharma, enabling better *in vitro* modeling to boost neurological research and drug discovery.

axoCells Sensory Neurons power NETRI microfluidics platforms

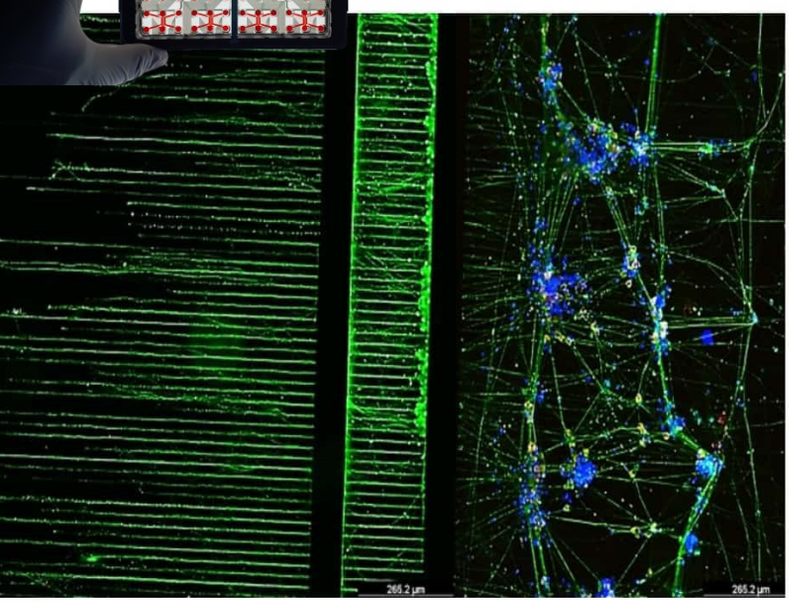
axoCells human iPSC-derived sensory neurons can power advanced in vitro platforms for models of pain, skin, and peripheral nervous system function.

Below they are growing on the NETRI NeoBento™ microfluidics platform, forming mature networks with neurite extension through the platform grooves (**fig. 7**).

With over a decade in the iPSC industry, we've developed the expertise to manufacture **high-quality, reproducible iPSC-derived cells at scale**, available to platform providers as the axoCells offering. axoCells are a **critical quality component** for platform providers looking to utilize iPSCs, and we're currently partnering with a growing list of providers looking to have their platforms powered by axoCells.



NETRI NeuroFluidics sensory neuron kit powered by axoCells Sensory Neurons



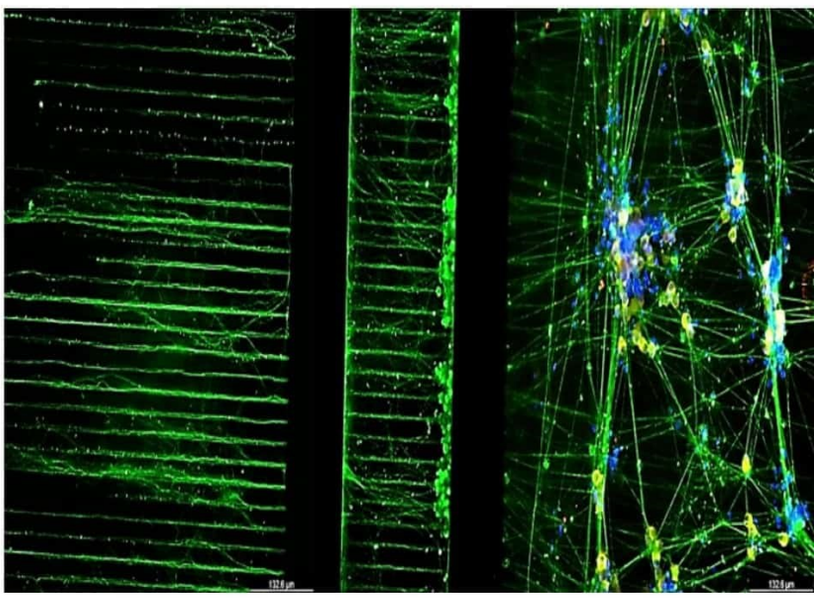


Figure 7. axoCells Sensory Neurons growing on the NETRI NeoBento™ microfluidics platform. Image at 10x and 20x magnification, staining for TUJ-1 (neuronal marker, green), $Na_V1.7$ (a key sensory neuron ion channel, yellow), and DAPI (nuclear stain, blue), demonstrating the presence of mature sensory neurons.

Powered by axoCells™

*"We will help to bolster the growing iPSC market through co-development of advanced iPSC platforms that rely on the **functional quality** and **consistency** of axoCells in combination with NETRI's high-relevance & high-throughput in vitro OOC models."*

Liam Taylor, CEO Axol Bioscience

axoCells Human iPSC-Derived Sensory Neurons

Supplied as Neural Stem Cells and a protocol to mature into final striatal neurons in **21 days**

Cells & Kits

Product Name	Cells only code / 1 vial	Quantity* / per vial	Kit** code
axoCells™ Human iPSC-Derived Neural Stem Cells, new-born male donor, ≥3.2 million cells	ax0555	≥1.5 million cells	-
axoCells™ Human iPSC-Derived Sensory Neurons, male donor, ≥0.5 million cells	ax0055	≥0.5 million cells	**ax0157**

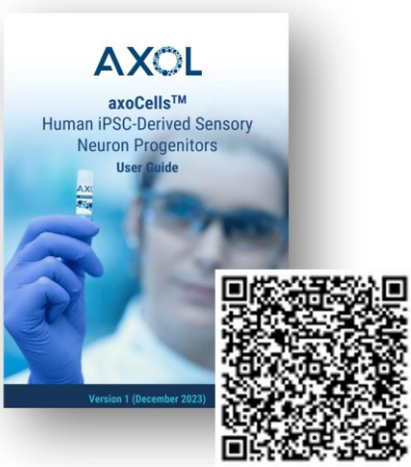
*Number of viable cells post thaw
**Kit contains cells and one of each item listed in the media and supplements table

Media and Supplements

Product Name	Product code	Quantity
axoCells™ Sensory Neuron Maintenance Media, 250 ml	ax0060	250 ml
axoCells™ Sensory Neuron Maximizer Supplement, 1 ml	ax0058	1 ml
axoCells™ Neural Plating Media, 30ml	ax0033	30 ml
axoCells™ Human GDNF Supplement, 10µg	ax139855	10 µg
axoCells™ Human BDNF Supplement, 10 µg	ax139800	10 µg
axoCells™ Human Beta-NGF Supplement, 20 µg	ax139789	20 µg
axoCells™ Human NT-3 Supplement, 10 µg	ax139811	10 µg

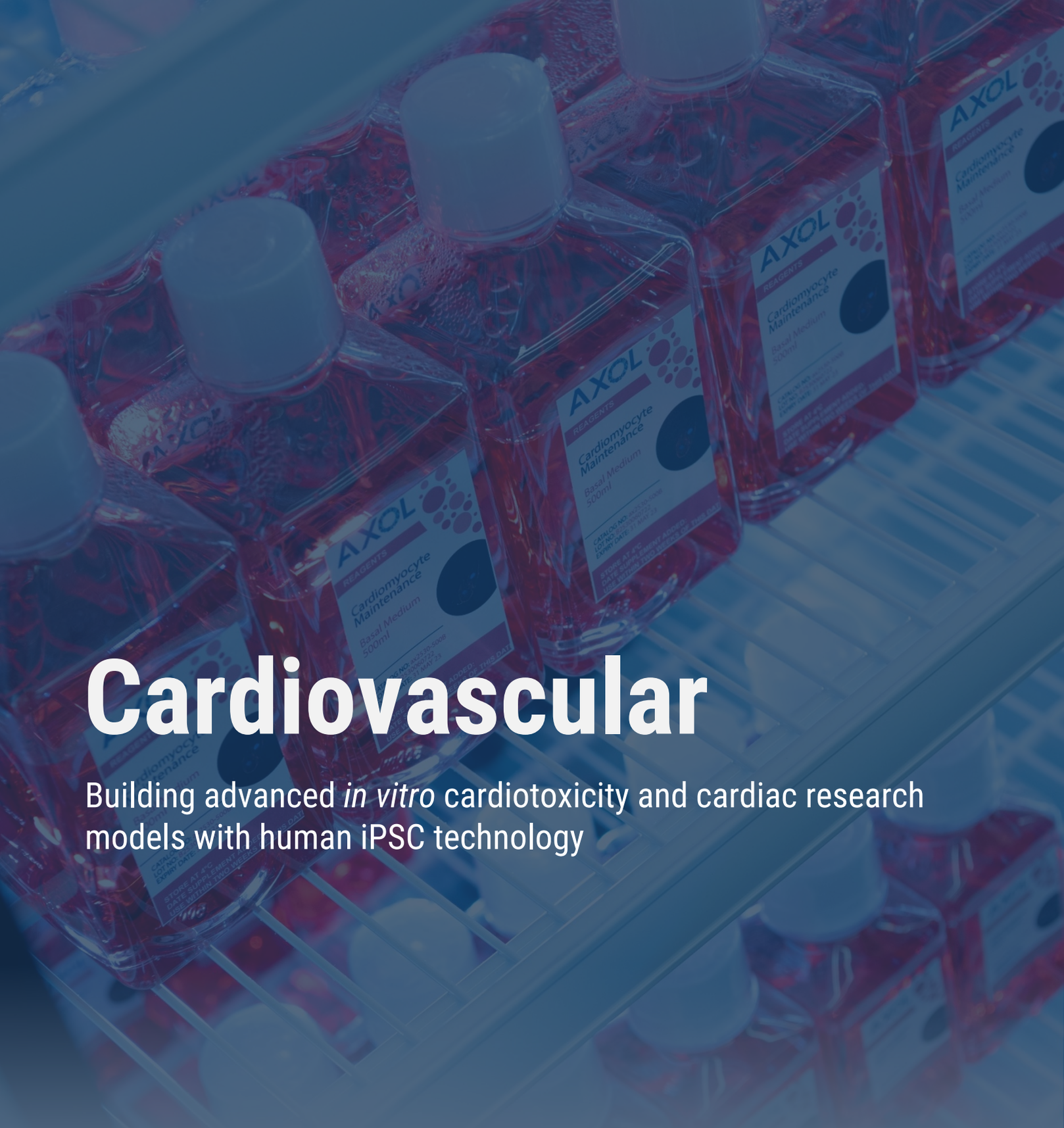
Additional third-party components may be required. Please refer to protocol for full list.

User Protocol



Can't see exactly what you need?

We can perform custom differentiation runs from either our lines or your lines. Please ask us about our 'made-to-order' service.



Cardiovascular

Building advanced *in vitro* cardiotoxicity and cardiac research models with human iPSC technology

Cardiovascular

iPSC-derived cardiac cells for research and cardiotoxicity screening

- Atrial cardiomyocytes
- Ventricular cardiomyocytes

Cardiotoxicity is responsible for one-third of pharmaceutical regulatory clearance failures, placing it amongst the biggest challenges in drug development[1]. Cardiac safety pharmacology has encountered well-characterized challenges, with Torsades de Pointes (TdP) and other fatal arrhythmias responsible for 14 major drug withdrawals [2,3].

To screen for this, a range of cardiotoxicity models have been used, including *ex vivo* and *in vivo* (animal) models, primary cell models (using cells taken directly from humans or animals), and *in vitro* immortalized cells expressing certain ion channels critical for cardiac function.

While these models have added to our understanding, there is still a **translational gap** between the "bench" and the "clinic", highlighting the need for more **physiologically-relevant model systems** that can better translate to humans[4].

This is where **human induced pluripotent stem cells (iPSCs)** hold great promise. Derived from the reprogramming of human donor material, iPSCs can be differentiated into cardiac cells and used to build *in vitro* models. And because they retain the donor characteristics (including disease mutations, complex ion channel activity and functional performance) they can offer a more **human-relevant model** for research and cardiotoxicity screening.

At Axol Bioscience, we've been working with iPSCs in a **quality-focused environment** for over a decade and have developed a deep understanding of the challenges of this space. Our **ISSCR-compliant** quality management system drives consistency and quality in iPSC products manufactured at our **ISO 9001-accredited** production facility.

With the **FDA Modernization Act 2.0** driving greater adoption of iPSC technology, we have engaged with the **Health and Environmental Sciences Institute (HESI)** and conducted external validation of our axoCells™ iPSC-derived ventricular cardiomyocytes against the **Comprehensive *in vitro* Pro-arrhythmia Assay (CiPA)** compound panel, demonstrating their value in cardiotoxicity models. We believe iPSCs will be transformative for cardiotoxicity and research, so we will continue to drive better quality standards to unlock the benefits of iPSC technology for researchers.

1 Grafton, F et al. doi: https://doi.org/10.7554/eLife.68714
2 DiMasi JA et al. doi: https://doi.org/10.1016/j.jhealeco.2016.01.012.
3 Blinova K et al. doi: https://doi.org/10.1016/j.celrep.2018.08.079.
4 Pognan, Fet al. doi: https://doi.org/10.1038/s41573-022-00633-x

Read more about the CiPA validation of our axoCells ventricular cardiomyocytes


APPLICATION NOTE

in vitro cardiotoxicity:
External validation of axoCells™ ventricular cardiomyocytes against CiPA cardiotoxicity compound panel

Background

Read more about the chamber-specific differences between our axoCells atrial and ventricular cardiomyocytes


APPLICATION NOTE

Functional and pharmacological differences between the contractility of axoCells™ iPSC-derived atrial and ventricular cardiomyocytes assessed on the FLEXcyte 96

axoCells™ Ventricular Cardiomyocytes

Ventricular cardiomyocytes represent those found in the ventricles of the human heart. They are frequently used to fuel ***in vitro*** **cardiotoxicity models** to assess drug safety and for **cardiac research**. They can also be used in **co-culture** with other cells (including atrial cardiomyocytes).

Spontaneously beat **3 days** post-thaw and assay-ready in just **7 days**

Validated against all **28 CiPA compounds**

Demonstrate **functional responses** in a **range of assay formats** including patch clamp, electrophysiology and voltage-sensitive dyes

Phenotypic characterization

We have conducted extensive characterization of our axoCells ventricular cardiomyocytes including morphology, immunocytochemistry (**fig.1**) and RNA sequencing, to assess their utility in iPSC-based cardiotoxicity models.

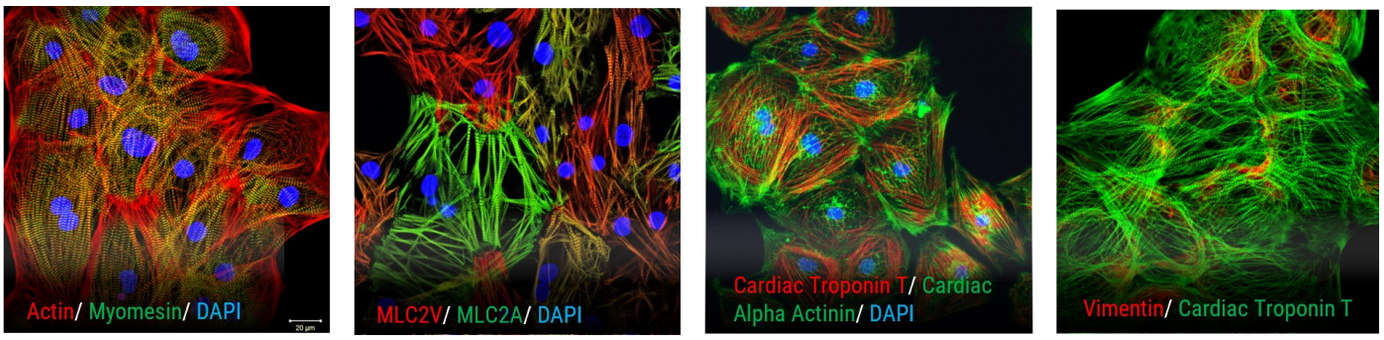


Figure 1. Immunocytochemistry of axoCells ventricular cardiomyocytes stained for key markers. Key morphological features are also demonstrated including sarcomeric alignment. Magnification 63x for images 1,2 and 4; 40x for image 3. Scale bar = 20 µm.

Functional relevance

We have validated the functional performance of our axoCells ventricular cardiomyocytes in a range of assays, including patch clamp (**fig. 2**), electrophysiology (**fig. 3**) and using voltage-sensitive dyes with the **Comprehensive *in vitro* Pro-arrhythmic Assay (CiPA) panel**. We have also demonstrated the chamber-specificity of our axoCells atrial and ventricular cardiomyocytes.

axoCells Ventricular Cardiomyocytes

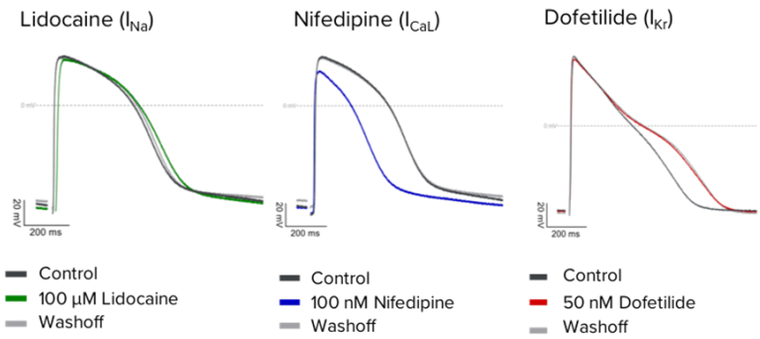


Figure 2. axoCells human iPSC-derived ventricular cardiomyocytes express the core cardiac ion channels I_{Na}, $I_{Ca,L}$ and I_{Kr}. Here we present representative patch clamp traces of evoked action potentials recorded under control conditions (grey) and in the presence of 100 µM Lidocaine (green), 100 nM Nifedipine (blue) or 50 nM Dofetilide (red), which show expected effects on action potential amplitude and duration.

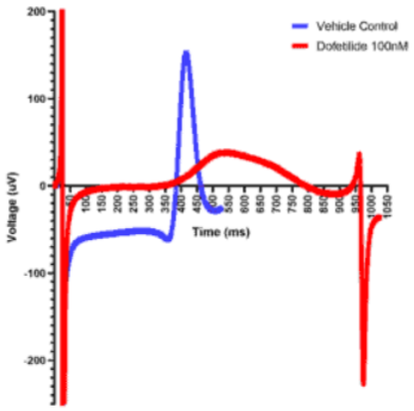


Figure 3. Multi-electrode array field action potential waveform of axoCells iPSC-derived ventricular cardiomyocytes. This demonstrates the effect of dofetilide (100nM, red) versus control (blue), where dofetilide causes prolonged field potential duration and after-depolarization, an expected response given its action as a hERG blocker.

Cells & Kits

Product Name	Cells only code / 1 vial	Quantity* / per vial	Kit** code
axoCells™ Human iPSC-Derived Ventricular Cardiomyocytes, male donor, ≥1 million cells	ax2508	≥1 million cells	**ax2500**

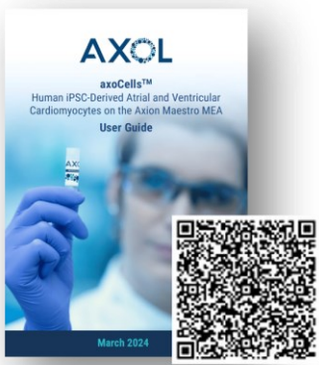
*Number of viable cells post thaw
**Kit contains cells and one of each item listed in the media and supplements table

Media and Supplements

Product Name	Product code	Quantity
axoCells™ Cardiomyocyte Maintenance Media, 500 ml	ax2530-500	500 ml
axoCells™ Fibronectin Coating, 100 µl	ax0050 (100 µL)	100 µl

Additional third-party components may be required. Please refer to protocol for full list.

User Protocols



axoCells™ Atrial Cardiomyocytes

Atrial cardiomyocytes represent those found in the atria of the human heart. They are frequently used in ***in vitro*** **cardiotoxicity models** to assess drug safety models and to support testing of irregular and abnormally fast heart rates (including atrial fibrillation). They can also be used in **co-culture** with other cells (including ventricular cardiomyocytes).

Spontaneously beat **3 days** post-thaw and assay-ready in just **7 days**

No evidence of endogenous arrhythmias

Demonstrate **functional response** to atrial-specific compounds including **carbachol**

Phenotypic characterization

We have conducted extensive characterization of our axoCells™ atrial cardiomyocytes including morphology, immunocytochemistry (**fig. 1**) and RNA sequencing, to assess their utility in iPSC-based cardiotoxicity models.

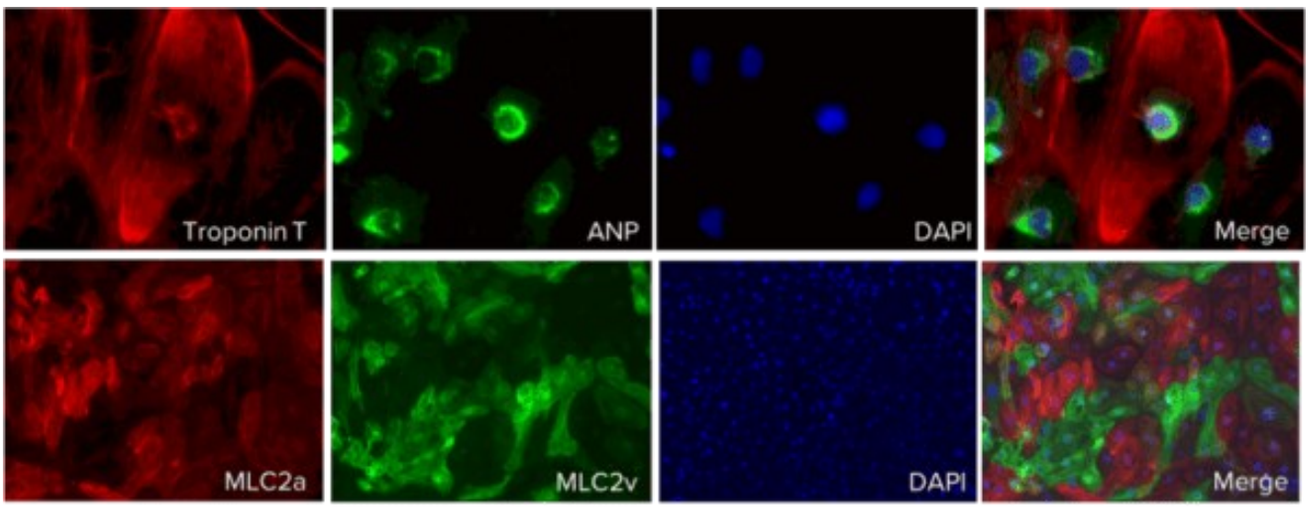


Figure 1. Immunocytochemistry demonstrating expression of key cardiac- and atrial-specific markers troponin T, atrial myosin light chain 2 (MLC2a) and atrial natriuretic peptide (ANP). Troponin T staining (red) confirmed the presence of cardiac myocytes, ANP is specifically secreted by atrial myocytes upon atrial stretching and MLC2a facilitates cardiac contractility. The nuclear marker DAPI was used as a counterstain.

Functional relevance

We have validated the functional performance of our axoCells atrial cardiomyocytes in a range of assays, including patch clamp (**fig. 2**), electrophysiology and contractility on the InnoVitro FLEXcyte 96 (**fig. 3**).

axoCells Atrial Cardiomyocytes

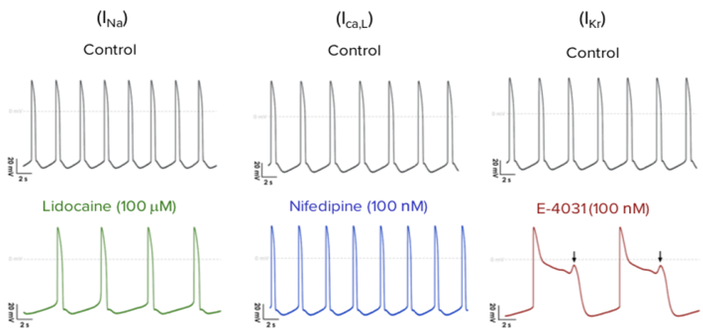
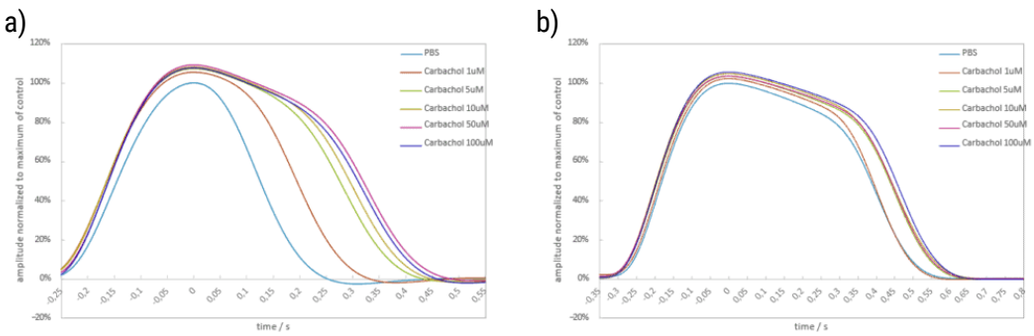


Figure 2. axoCells atrial cardiomyocytes express functional core cardiac ion channels, I_{Na}, $I_{Ca,L}$ and I_{Kr}.
Lidocaine (I_{Na}), Nifedipine ($I_{Ca,L}$), and E-4031 (I_{Kr}) were used to characterize the activity of core cardiac currents during action potentials. Each trace shows representative spontaneous action potentials under control conditions (grey) and in the presence of 100 µM Lidocaine (green), 100 nM Nifedipine (blue), and 100 nM E-4031 (red). Early afterdepolarizations were observed after application of the hERG blocker E-4031 (arrow), indicative of arrhythmic activity. Work in collaboration with Metrion Biosciences.

Figure 3. Chamber-specific pharmacological response.
The effect of carbachol (an atrial-specific activator of I_{KAch}) on the contractility of axoCells atrial (**3a**) and ventricular (**3b**) cardiomyocytes, as measured on the InnoVitro FLEXcyte 96. As expected, carbachol has a marked effect on action potential duration in atrial cardiomyocytes but not in ventricular cardiomyocytes.



Cells & Kits

Product Name	Cells only code / 1 vial	Quantity* / per vial	Kit** code
axoCells™ Human iPSC-Derived Atrial Cardiomyocytes, male donor, ≥1 million cells	ax2518	≥1 million cells	**ax2510**

*Number of viable cells post thaw
**Kit contains cells and one of each item listed in the media and supplements table

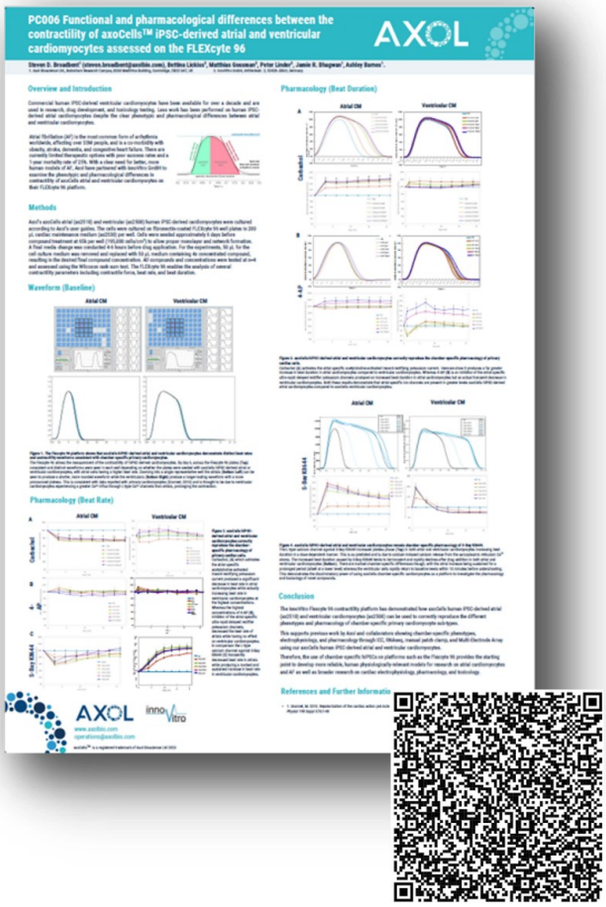
Media and Supplements

Product Name	Product code	Quantity
axoCells™ Cardiomyocyte Maintenance Media, 500 ml	ax2530-500	500 ml
axoCells™ Fibronectin Coating, 100 µl	ax0050 (100 µL)	100 µl

Additional third-party components may be required. Please refer to protocol for full list.

User Protocols

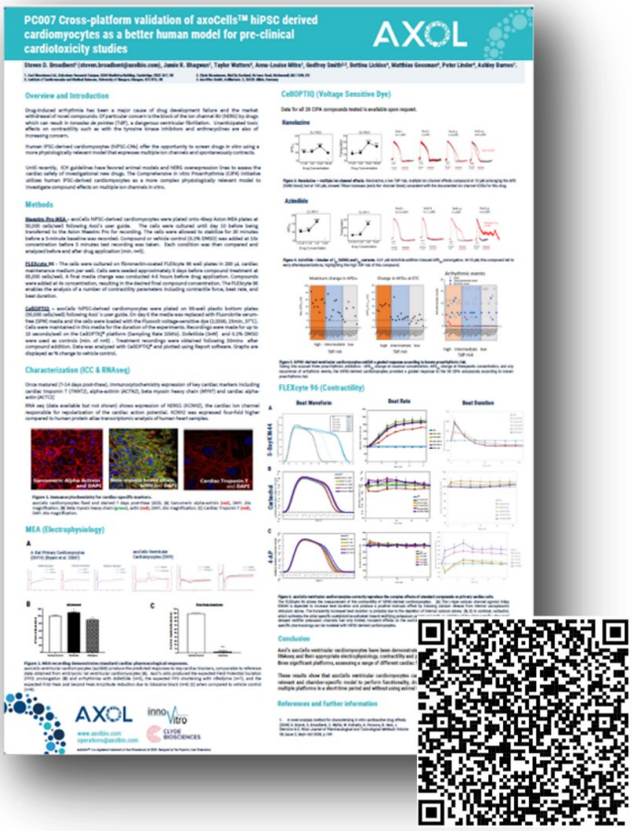
Recent posters



Functional and pharmacological differences between the contractility of axoCells™ iPSC-derived atrial and ventricular cardiomyocytes assessed on the FLEXcyte 96

Here we demonstrate that our axoCells iPSC-derived isogenic atrial and ventricular cardiomyocytes show distinct physiologically relevant phenotypes in contractility and pharmacology assays. This demonstrates the suitability of these cells for chamber-specific *in vitro* cardiac models.

Scan the QR code to access the poster.



Cross-platform validation of axoCells™ hiPSC-derived cardiomyocytes as a better human model for pre-clinical cardiotoxicity studies

Here we outline the performance of our axoCells ventricular cardiomyocytes across three significant platforms that assess marker expression, electrophysiology, contractility and pharmacology. This demonstrates the value of axoCells ventricular cardiomyocytes for robust, chamber-specific *in vitro* models for cardiotoxicity screening and drug discovery.

Scan the QR code to access the poster.

About Axol Bioscience

Like you, we believe that having more human-relevant disease models will **expand scientific knowledge** and **de-risk drug development**. We use human iPSCs to achieve this and have been doing so for over a decade.

We take cells from patient and healthy donors and, using our **leading iPSC technology**, work with researchers to build physiologically-relevant *in vitro* models. We have a special focus on neurodegenerative diseases (like Alzheimer's Disease) as well as cardiotoxicity to promote drug safety.

Our customers benefit from our **extensive experience**, meaning we can do the scientific "heavy lifting" to unlock the benefits of iPSC technology. That ultimately means **more confidence** in the data outputs of advanced *in vitro* models, along with **better insights** and **reduced costs**.

So we ask you: iPSCs? What can we do to help?

www.axolbio.com