



# SZABO SCANDIC

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## Produktinformation



Forschungsprodukte & Biochemikalien



Zellkultur & Verbrauchsmaterial



Diagnostik & molekulare Diagnostik



Laborgeräte & Service

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See the following pages for more information!



### Lieferung & Zahlungsart

siehe unsere [Liefer- und Versandbedingungen](#)

### Zuschläge

- Mindermengenzuschlag
- Trockeneiszuschlag
- Gefahrgutzuschlag
- Expressversand

### SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

[mail@szabo-scandic.com](mailto:mail@szabo-scandic.com)

[www.szabo-scandic.com](http://www.szabo-scandic.com)

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 



6042 Cornerstone Court W, Ste B  
San Diego, CA 92121  
Tel: 1.858.202.1401  
Fax: 1.858.481.8694  
Email: [info@bpsbioscience.com](mailto:info@bpsbioscience.com)

**Data Sheet**  
**FGFR2 Assay Kit**  
Catalog #79804  
96 Reactions

**BACKGROUND:** FGFR2 (Fibroblast Growth Factor Receptor 2) is a member of a family of tyrosine kinases involved in many pathways that play a significant role in cancer. Amplification or activation of FGFR2 has been reported in breast and gastric cancers, while FGFR2 mutations have been observed in endometrial and breast cancers. Mutations in FGFR2 are also associated with bone development disorders including Pfeiffer Syndrome and Crouzon Syndrome.

**DESCRIPTION:** The *FGFR2 Assay Kit* is designed to measure FGFR2 activity for screening and profiling applications using Kinase-Glo<sup>®</sup> MAX as a detection reagent. The *FGFR2 Assay Kit* comes in a convenient 96-well format, with enough purified recombinant FGFR2, Protein Tyrosine Kinase Substrate (Poly-Glu,Tyr 4:1), ATP, and kinase assay buffer for 96 enzyme reactions.

**COMPONENTS:**

| Catalog # | Reagent   | Amount | Storage |   |
|-----------|---|--------|---------|---|
| 40211     | FGFR2, GST-Tag  | 5 µg   | -80°C   | <b>Avoid multiple freeze/thaw cycles!</b> |
| 79334     | 5x Kinase assay buffer  | 1.5 ml | -20°C   |   |
| 79686     | ATP (500 µM)  | 100 µl | -20°C   |   |
| 40217     | Protein Tyrosine Kinase Substrate (Poly-Glu,Tyr 4:1) (10 mg/ml) | 100 µl | -20°C   |   |
| 79696     | 96-well plate, white  | 1      | RT      |   |

**MATERIALS OR INSTRUMENTS REQUIRED BUT NOT SUPPLIED:**

Kinase-Glo MAX (Promega #V6071)  
Dithiothreitol (DTT, 0.5 M; optional)  
Microplate reader capable of reading luminescence  
Adjustable micropipettor and sterile tips  
30°C incubator

**APPLICATIONS:** Useful for studying enzyme kinetics and screening small molecular inhibitors for drug discovery and HTS applications.

**STABILITY:** Up to 6 months when stored as recommended.

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#### REFERENCE:

1. Rutland, P., *et al.* 1995. "Identical mutations in the FGFR2 gene cause both Pfeiffer and Crouzon syndrome phenotypes." *Nature Genetics* **9(2)**: 173.
2. Xie, L., *et al.* 2013. "FGFR2 gene amplification in gastric cancer predicts sensitivity to the selective FGFR inhibitor AZD4547." *Clinical Cancer Research* **9(2)**: 2572-2583.

#### ASSAY PROTOCOL:

**All samples and controls should be tested in duplicate.**

- 1) Thaw **5x Kinase assay buffer**, **ATP (500  $\mu$ M)**, and **Protein Tyrosine Kinase Substrate (Poly-Glu,Tyr 4:1)**.  
(Optional: If desired, add 30  $\mu$ l of 0.5 M DTT to **5x Kinase assay buffer**).
- 2) Prepare the master mixture (25  $\mu$ l per well): N wells x (10  $\mu$ l **5x Kinase assay buffer** + 1  $\mu$ l **ATP (500  $\mu$ M)** + 1  $\mu$ l **Protein Tyrosine Kinase Substrate (Poly-Glu,Tyr 4:1)** + 13  $\mu$ l distilled water). Add 25  $\mu$ l to every well.

|                                      | Positive Control | Test Inhibitor | Blank      |
|--------------------------------------|------------------|----------------|------------|
| 5x Kinase assay buffer               | 10 $\mu$ l       | 10 $\mu$ l     | 10 $\mu$ l |
| ATP (500 $\mu$ M)                    | 1 $\mu$ l        | 1 $\mu$ l      | 1 $\mu$ l  |
| Poly-Glu,Tyr(10 mg/ml)               | 1 $\mu$ l        | 1 $\mu$ l      | 1 $\mu$ l  |
| Water                                | 13 $\mu$ l       | 13 $\mu$ l     | 13 $\mu$ l |
| Test Inhibitor                       | -                | 5 $\mu$ l      | -          |
| 10% DMSO in Water (inhibitor buffer) | 5 $\mu$ l        | -              | 5 $\mu$ l  |
| 1x Kinase buffer                     | -                | -              | 20 $\mu$ l |
| FGFR2, GST-tag (2.5 ng/ $\mu$ l)     | 20 $\mu$ l       | 20 $\mu$ l     | -          |
| Total                                | 50 $\mu$ l       | 50 $\mu$ l     | 50 $\mu$ l |

- 3) Add 5  $\mu$ l of Inhibitor solution of each well labeled as "Test Inhibitor." For the "Positive Control" and "Blank," add 5  $\mu$ l of the same solution without inhibitor (Inhibitor buffer), usually 10% DMSO in water

*Note: Final DMSO concentration must be  $\leq$ 1%. Higher DMSO levels can significantly decrease the enzyme activity. For example, to test an inhibitor dissolved in 100% DMSO at 10  $\mu$ M, dilute 1 mM inhibitor with water to make a 100  $\mu$ M inhibitor in 10% DMSO(aq). Then, add 5  $\mu$ l of the 100  $\mu$ M solution into the 50  $\mu$ l assay to make a 1% DMSO concentration in the final reaction mixture.*

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- 4) Prepare 3 ml of **1x Kinase assay buffer** by mixing 600  $\mu$ l of **5x Kinase assay buffer** with 2400  $\mu$ l water. 3 ml of **1x Kinase assay buffer** is sufficient for 100 reactions.
- 5) To the wells designated as "Blank," add 20  $\mu$ l of **1x Kinase assay buffer**.
- 6) Thaw **FGFR2, GST-Tag** on ice. Upon first thaw, briefly spin tube containing enzyme to recover full content of the tube. Calculate the amount of **FGFR2, GST-Tag** required for the assay and dilute enzyme to 1 ng/ $\mu$ l with **1x Kinase assay buffer**. Store remaining undiluted enzyme in aliquots at  $-80^{\circ}\text{C}$ . *Note: FGFR2, GST-Tag is sensitive to freeze/thaw cycles. Avoid multiple freeze/thaw cycles. Do not re-use thawed aliquots or diluted enzyme.*
- 7) Initiate reaction by adding 20  $\mu$ l of diluted **FGFR2, GST-Tag** to the wells designated "Positive Control" and "Test Inhibitor Control." Incubate at  $30^{\circ}\text{C}$  for 45 minutes.
- 8) Thaw Kinase-Glo Max reagent.
- 9) After the 45 minute reaction, add 50  $\mu$ l of Kinase-Glo Max reagent to each well. Cover plate with aluminum foil and incubate the plate at room temperature for 15 minutes.
- 10) Measure luminescence using the microplate reader. Value of "Blank" reading should be subtracted from all other measurements.

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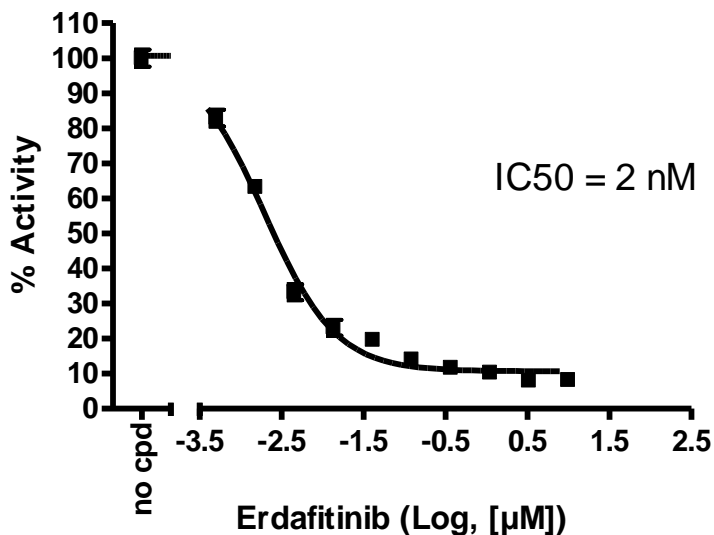
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**Example of Assay Results:**

**FGFR2, GST-Tag Activity**



Inhibition of FGFR2, GST-Tag by Erdafitinib, measured using the FGFR2 assay kit (BPS Bioscience #79804). *Data shown is lot-specific. For lot-specific information, please contact BPS Bioscience, Inc. at [info@bpsbioscience.com](mailto:info@bpsbioscience.com)*

**RELATED PRODUCTS:**

| <u>Product Name</u>                                     | <u>Catalog #</u> | <u>Size</u> |
|---|------------------|-------------|
| FGFR2, GST-tag  | 40211            | 10 µg       |
| 5x Kinase assay buffer                                  | 79334            | 10 ml       |
| ATP (500 µM)  | 79686            | 200 µl      |
| Protein Tyrosine Kinase Substrate<br>(poly-Glu,Tyr 4:1) | 40217            | 1 mg        |
| FGFR1(FLT2), GST-tag                                    | 40210            | 10 µg       |
| FGFR1 (V561M), GST-tag                                  | 40209            | 10 µg       |
| FGFR3 (CD333), GST-tag                                  | 40212            | 10 µg       |
| FGFR3 (V443M), His-Tag                                  | 100140           | 10 µg       |
| FGFR3 (L496V), His-Tag                                  | 100142           | 10 µg       |
| FGFR3 (V443L), His-Tag                                  | 100138           | 10 µg       |
| FGFR4, GST-tag  | 40213            | 10 µg       |
| FGFR1(FLT2), GST-tag                                    | 40210            | 10 µg       |
| FGFR1(FLT2), GST-tag                                    | 40210            | 10 µg       |
| FGFR1(FLT2), GST-tag                                    | 40210            | 10 µg       |
| FGFR1(FLT2), GST-tag                                    | 40210            | 10 µg       |

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