What makes G+® Graphene Plus a safe choice:

Due to various production processes and distinct physical/chemical characteristics graphene materials differ from one another. Here below we explain why among different graphene materials G+® is the safest choice

- ➔ G+® Graphene Plus is obtained by purely physical treatments of natural graphite, thus avoiding any chemical treatments with organic solvents or acids, and **just exploiting water, temperature and pressure** to reduce the graphite thickness to the nanometric level. Many competitors use chemicals and often also nanometals within the production process. These additives cannot be completely removed from the final graphene enhanced product, thus potentially representing a health hazard.

- ➔ G+® Graphene Plus is made of graphene nanoplatelets with a lateral dimension (X-Y) in the micrometer range. **Only the thickness (Z) is nanometric**. This is a key feature to properly categorize graphene morphology. Many competitors produce graphene particles which are nanometric in all the three dimensions (lateral sizes X-Y and thickness Z): such particles are strictly defined as nanoparticles and behave differently than particles with just one dimension in the nanometric range, especially in terms of biological interactions.

- ➔ Directa Plus has always considered the health and safety of all stakeholders and the environmental protection as top priorities. Since our company was established, we implement a proactive approach to health, safety and environmental protection by monitoring our production process and products: continuously searching for new testing protocols and aligning with the latest research on nanomaterials and international regulation.

- ➔ **Since 2013, we collected 43 certificates reporting the absence of negative impacts on biological systems and on environment applications.** Hereafter we bring evidence of all the tests and reports we collected on HS&EP analysis.

Please note: PURE G+® is the commercial name of Pristine Graphene Nanoplatelets (GNPs) powder; GRAFYTEX SP9 is a printing paste with graphene (GNPs) and GRAFYPAD G+® is a graphene (GNPs) additive for impregnation, for textile application. GRAFYSORBER G+® is super expanded graphite, for environmental applications.

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1. REACH REGISTRATION

Directa Plus, on April 2017, obtained the REACH registration for their materials produced along the production line in the graphite consortium. This certification entitles the company to fabricate expanded graphite and graphene nanoplatelets, derived from graphite, for a quantity between 10 and 100 tons.

In October 2018, GRAFYTEX SP9 product was declared compliant with the European Regulation 1907/2006 / EC (REACH).

In May 2019, according to the new regulation, Directa Plus applied and successfully completed the REACH registration of its pristine graphene nanoplatelets in the graphene consortium (EC number: 801-282-5 | CAS number: 1034343-98-0).

2. HEALTH PROTECTION – Quantification of the concentration of nanoparticles in the Graphene Factory

Directa Plus is the first company among producers and suppliers of graphene-based products to conduct and promote extensive research on the impact of nanomaterials on human health and in the workplace and to advocate the adoption of stringent safety procedures.

As part of this, Directa Plus was a key participant in the MULAN program (MULtilevel Approach to the study of Nanomaterials Health and Safety), which ran from October 2013 to February 2016.

Under the MULAN program, Directa Plus evaluated the following parameters:

- Concentration of nanoparticles in urban environments (outdoor and indoor) and in occupational settings
- Exposure to nanoparticles of people working with nanomaterials

The results of this analysis showed a negligible concentration of nanoparticles in the workplace. The results have been published in "Engineered nanomaterials exposure in the production of graphene" in Aerosol Science and Technology.

We continue to evaluate the production plant in order to regularly verify particulate levels in the workplace. In fact, we repeated the same evaluation in 2018 and we saw that the total amount of powder or nanoparticles in the Graphene Factory was below the instrumental quantification limit and also far below the limits imposed by the American Conference of Governmental Industries of Hygienists (ACGIH)

3. HEALTH PROTECTION – Quantification of the concentration of airborne Ultrafine Particle (UFP - diameter <100 nm) and Particulate Matter (PM - coarse PM, diameter > 1 µm and fine PM, diameter < 1µm) under chemical hood after handling of Graphene Nanoplatelets (GNPs)

The goal of this research was to examine the persistence of GNPs at the end of handling. Consequently, the equipment conditions involving less GNPs dispersion in the hood compartment and in the laboratory environment, and less GNPs exposure of the operator involved in the handling of the material, have been assessed.

During the handling of GNPs, a small increment of PM > 1 µm is observed but the persistence of GNPs at the end of handling is quantified in less than 5 minutes.

The concentration levels of GNPs observed are, however, always particularly modest and below the Nano Reference Values (NRVs) relative to the typology to which our GNPs belong.

4. Toxicology screening on G+ products PURE G+ and GRAFYSORBER _ *in vitro* tests

Directa Plus performed and completed independently a consistent series of toxicology screening tests to certify the non-toxicity and non-cytotoxicity of G+ products PURE G+ and GRAFYSORBER.
All the analyses have been performed following international standard method.
The toxicology analysis performed on Directa Plus products included the following seven tests:

1- **CITOTOXIC POTENTIAL**: G+ products were classified as **NOT TOXIC** - UNCLASSIFIED for acute oral toxicity
2- **SKIN IRRITATION**: G+ products were classified as **NOT IRRITANT**
3- **SKIN CORROSION**: G+ products were classified as **NOT SKIN CORROSIVE**
4- **EYE IRRITATION**: G+ products were classified as **NOT EYE IRRITANT** (UN GHS no category)
5- **PRO-SENSITISING POTENTIAL:** G+ products **didn't show any detectable pro-sensitising effects**
6- **REPRODUCTION TOXICITY**: G+ products were classified as **NOT TOXIC FOR REPRODUCTION**
7- **GENOTOXICITY**: G+ products were classified as **NOT GENOTOXIC**

5. Acute inhalation toxicity of G+ product PURE G+ _ *in vivo* test

Directa Plus performed an *in vivo* test to evaluate the acute inhalation toxicity of the test item PURE G+ in male and female Sprague Dawley rats by the traditional protocol described in the OECD Guideline Nº 403 (international standard method): Acute Inhalation Toxicity.
This method provides lethality data of the test item and allows its classification according to the Globally Harmonized System (GHS) of Classification and Labelling of Chemicals, that classifies materials in 5 Categories, where the Category 1 means "fatal if inhaled", while the Category 5 means "may be harmful if inhaled" (see table 1 below).
At the end of the test, and based on the GHS classification criteria, PURE G + can be considered to be under human health hazard Category 4 or higher (Category 5).

	Category 1	Category 2	Category 3	Category 4	Category 5
--Inhalation *see Note*	Fatal if inhaled	Fatal if inhaled	Toxic if inhaled	Harmful if inhaled	May be harmful if inhaled

Table 1: GHS classification table

PURE G+ was therefore put in the Categories indicating low inhalation toxicity.

6. Toxicology screening on G+ textile products _ *in vivo* tests

Directa Plus performed *in vivo* tests on human volunteers to evaluate toxicological aspects of G+ printed fabrics, G+ impregnated fabrics, G+ membranes and coated samples with G+.
Allergic skin tests, of different durations, have been carried out to evaluate potential skin irritation after contact with G+ fabrics, G+ coated fabric or G+ membranes.
Directa Plus textile products, at the end of the analysis, have been certified as:

- DERMATOLOGICALLY TESTED, after 48 hours of applications
- HYPOALLERGENIC, after three weeks of applications on volunteers with sensitive skin

7. ZDHC and MRSL for GRAFYTEX SP9 and GRAFYPAD G+

ZDHC stands for Zero Discharge of Hazardous Chemicals which is an organization dedicated to eliminating hazardous chemicals, and implementing sustainable chemicals in the leather, textile and synthetics sectors.

The ZDHC Program is a multi-stakeholder group which includes brands, value chain affiliates and associates, that work collaboratively to implement responsible chemical management practices.

One of the tools that ZDHC has developed to achieve their objective is the ZDHC MRSL. The ZDHC MRSL (Manufacturing Restricted Substance List) is a list of chemical substances banned from intentional use in facilities that process textile materials and trim parts in apparel and footwear. The ZDHC MRSL establishes acceptable concentration limits for substances in chemical formulations used within manufacturing facilities.

The limits are designed to eliminate the possibility of intentional use of listed substances. The intent of the ZDHC MRSL is to manage the input of chemicals to the suppliers and remove those hazardous substances from the manufacturing process.

Grafytex SP9 and GRAFYPAD G+ were tested for ZDHC MRSL and resulted compliant.

8. LCA for Grafysorber G+

This document aims to investigate and define the Life Cycle and the Environmental Impact of Grafysorber for oils and hydrocarbons removal from contaminated water. The first version of this Life Cycle Assessment was carried out as voluntary Directa Plus' decision and was approved by the European Committee inside GEnIuS Project Action (www.genius-project.com). The assessment aims to make a qualitative and quantitative evaluation of the environmental impacts coming from Grafysorber® production process and tries to make a qualitative and semi-quantitative evaluation of those deriving from the use of Grafysorber® as oil-adsorbent material.

9. Nanoparticles release from G+ printed and impregnated fabric, and from Surgical Grey Filter during abrasion test

Determination of particle release when an abrasion force is applied has been done by following the ISO 12947 standard for "Determination of the abrasion resistance of fabrics". We have identified and characterized particle release by real-time and static monitoring methods. IOM/SAFENANO's established practices for conducting occupational hygiene surveys are consistent with the OECD recommendations for the identification and assessment of emissions of airborne manufactured nanomaterials in the workplace, these principles have been adapted to measure and quantify release from Directa Plus product.

Under the conditions used, the release of graphene nanoplatelets from G+ fabrics was, at best, not observed, and at worst, at low levels.

10. Nanoparticles release from G+ printed fabric and G+ impregnated cotton in biological fluids (bioelution)

Determination of release of free graphene from the graphene paste additive, testing has been conducted on the base textile with and without graphene additives in triplicate at each time point using a standard IOM bioelution test, with reference to PD ISO/TR 19057:2017 standard, using a simulated sweat (SSW) solution.

Under the conditions used, the release of graphene nanoplatelets was, at best, not observed, and at worst, at low levels.

11. ECOPASSPORT by OEKO-TEX Certification of Grafytex G+ and for Grafypad G+

ECO PASSPORT by OEKO-TEX® is an independent certification system for chemicals, colorants and auxiliaries used in the textile and leather industry. During a multistep process, the testing institute analyses whether

each individual ingredient in the chemical product meets the statutory requirements and that it is not harmful to human health.

The textile chemicals certified in accordance with the ECO PASSPORT have been tested for harmful substances in critical concentrations as listed in the ECO PASSPORT standard.

G+ products GRAFYTEX SP9, GRAFITEX SP11 and GRAFYPAD G+ received the ECOPASSPORT by OEKOTEX certification.

12. Ecotoxicity test in fresh water on *P. subcapitata* seaweeds

The aim of the test is to determine the ecotoxic effects of GRAFYSORBER® G+ in biological systems. In particular, a growth inhibition test on the alga *Pseudokirchneriella subcapitata* in fresh water was conducted according to OECD 201 and the ISO8692 guidelines

The test was performed on the extracted filtered diluted solution of GRAFYSORBER® G+ at 0.063, 0.125,0.25, 0.5, 1 g/L, for a total period of 72 hours.

The sample extract showed a lack of ecotoxic potential in the applied experimental model.

In compliance with OECD 201 validity criteria the product extract inducing the 50% alga growth inhibition is > 100 mg/l (EC50 % > 100mg/L).

13. Mucosal tollerability on alveolar epithelium (cell viability and inflammatory cytokine quantification) (PURE G+)

Evaluation of alveolar epithelium tolerance of PURE G+ by means of the evaluation of the viability of the tissue used in the test and the release of TNF-alpha (proinflammatory acute cytokine) following the treatment with the product under exam.

The results of cell viability test in tissues treated with Pure G+ for 6 or 24 hours classify Pure G+ as NON IRRITANT product. The assay of the pro-inflammatory cytokine TNFalpha shows that tissues treated with PURE G + did not show statistically significant changes in the release of TNFalpha compared to CTR- ($p > 0.05$ vs CTR-) and therefore no inflammatory reactivity was highlighted.

14. Skin Absorption: In Vitro Method (PURE G+)

The purpose of the test was the evaluation of the skin absorption potential of the graphene Pure G+ through an in vitro reconstructed human epidermis. The penetration study was performed following the general principles of OECD 428.

Pure G+ was applied on tissue surface for 6 and 24 hours. At the end of the experimental time, the calculation of the quantity of graphene penetrated through the epidermis was performed.

Collected experimental data does not show any absorption potential for G+: no statistical differences were recorded among the control conditions and treated ones.

15. Ecotoxicity test in sea water on alga P.tricornutum

The aim of the test is to determine the ecotoxic effects of GRAFYSORBER® G+ in biological systems. In particular, a growth inhibition test on the alga Phaeodactylum tricornutum was conducted according to ISO10253 guidelines.

The test was performed on the extracted filtered diluted solution of GRAFYSORBER® G+ at 1 g/L, for a total period of 72 hours.

The sample extract showed a lack of ecotoxic potential in the applied experimental model.

In compliance with ISO 10253:2016 validity criteria the product extract inducing the 50% alga growth inhibition is > 100 mg/l (EC50 % > 100mg/L).

16. Ecotoxicity test with *Daphnia magna*

The aim of the test is to determine the ecotoxic effects of GRAFYSORBER® G+, through the evaluation of the mortality induced in Daphnia magna crustacean after 24h of contact with G+.

The sample extracted showed a lack of ecotoxic potential, with a Daphnia mortality of 0%.

17. Ecotoxicity test with *Artemia franciscana*

The aim of the test is to determine the ecotoxic effects of GRAFYSORBER® G+, through the evaluation of the mortality induced in *Artemia franciscana*, a well-established model organism for marine ecotoxicological studies after 96h of contact with G+.

The sample extracted showed a lack of ecotoxic potential.

18. Italian Ministry of the Environment authorization for the use of Grafysorber G+ for the remediation of hydrocarbon contamination in sea

Grafysorber G+ received the recognition and the authorization for its use for the remediation of contamination from petroleum hydrocarbons according to the DD March 31, 2009 and subsequent amendments.

19. EPA authorization of usage of Grafysorber G+ in USA

Grafysorber G+ is identified by EPA as a sorbent material and consists solely of the materials listed in section 300.915(g)(1) of the NCP; its usage is so authorized in the USA.