# Welcome to RPS Online 1.0: the Users Guide

**RPS Online (the Resistant Procrustes Software webapp)** provides a flexible and user-friendly platform to perform a descriptive landmark-based resistant shape analysis of 2D and 3D configurations of landmarks.

This open source software has a well-designed graphical user interface that offers a friendly and customizable working environment. Its overall performance is reasonably fast and efficient despite the time complexity that is typical in resistant algorithms. RPS Online modular design enables a faster, easier and more efficient customisation of the different analyses.

### About this document

RPS Online: User Guide is the primary reference when using RPS Online, giving a brief overview for all of its functionalities.

#### **BRIEF OVERVIEW OF THE FUNCTIONALITIES**

## **Procrustes superimpositions**

To perform a landmark-based shape comparison of several objects (landmarks configurations), a common coordinate system is needed. The purpose of a superimposition method is to find an adequate common reference frame, since the resulting shape differences highly depend on the chosen alignment criterion. It is known that a useful superimposition of shapes is achieved when differences due to location, scale and orientation are filtered out; these are known as Procrustes superimpositions. Two versions of them are implemented in RPS Online: the novel resistant Procrustes superimposition (Torcida et al. 2014) and the well known least squares Procrustes superimposition.

## **Generalized resistant Procrustes superimposition (GrP)**

Intuitively, a resistant alignment of two configurations of landmarks is achieved when those landmarks exhibiting no variation are perfectly superimposed. Then, shape differences can be clearly exhibited through the lack of fit of the remaining landmarks.

Unlike the least squares Procrustes superimposition, the GrP does not minimize an explicit mathematical criterion. Instead, it uses a repeated medians calculation to obtain optimal alignment parameters. The repeated medians technique enjoys the highest breakdown value: this means that whenever (possibly large) shape differences are located in less than 50% of the landmarks, a perfect superimposition of the remaining (unchanged) majority is achieved. In this way, the method "resists" the deformation present in just a few landmarks and the resulting superimposition follows the main trend. (Whenever shape differences are located in more than 50% of the landmarks, their measurement and/or recognition always becomes ambiguous).

The GrP implementation is completely based on <u>Torcida et al. (2014)</u>. The algorithm iteratively matches every configuration of landmarks to a resistant consensus whose landmarks are, respectively, the 3D spatial or geometric medians of the corresponding landmarks.

Following a GrP, localized shape differences between configurations are typically more accurately depicted. This facilitates the biological understanding and interpretation of shape variation in most of the cases.

### **Generalized least-squares Procrustes superimposition (GIsP)**

The GIsP (or classical) Procrustes superimposition is the standard alignment criterion within Geometric Morphometrics. This method minimizes the sum of squared Euclidean distances between the Cartesian coordinates of corresponding superimposed landmarks, iteratively matching every configuration to an average consensus.

Unique alignment parameters are found in the process, and maximal agreement in a least squares sense is thus achieved. It is widely known that this method is highly misleading when localized shape variation is present, because it averages the lack of fit among all the landmarks.

### **Morphometric Distances**

Following a Procrustes superimposition, the exhibited shape differences between any pair of configurations of landmarks need to be accordingly measured. Pairwise distances are typically computed and stored in a distance matrix, which in turn can be used as an input for a subsequent ordination or to perform other multivariate analyses. For each superimposition method, RPS Online can compute a coherent distance matrix.

Following a GrP, a reasonable distance between two objects is given by the sum of the resulting non-squared Euclidean distances across corresponding landmarks (<u>Torcida et al. 2014</u>). This distance is available in <u>RPS Online</u>.

Additionally, a least squares Distance (IsD) can be also computed: following a GIsP, the magnitude of shape differences between any two configurations of landmarks is measured by the squared root of the resulting sum of squared Euclidean distances across corresponding landmarks (which is often named as Procrustes distance)

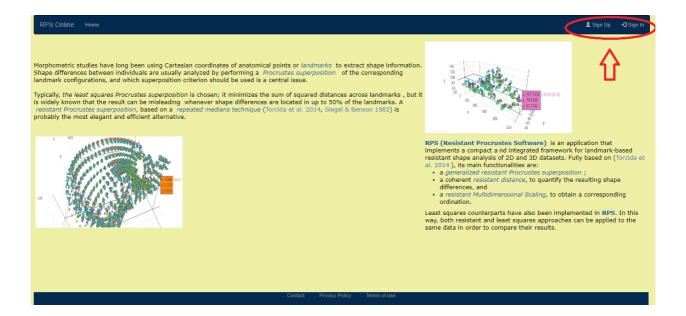
## **Ordinations**

When a Procrustes superimposition has been performed, a distance matrix is often computed to produce either a 2D or 3D graphical ordination which facilitates the visualization of the resulting shape differences. RPS Online implements the versatile Universal Multidimensional Scaling (UMDS) framework (Agarwal et al. 2010) which includes the classical least squares UMDS (IsUMDS) and a corresponding resistant UMDS (rUMDS).

### **USAGE**

#### **User Interface**

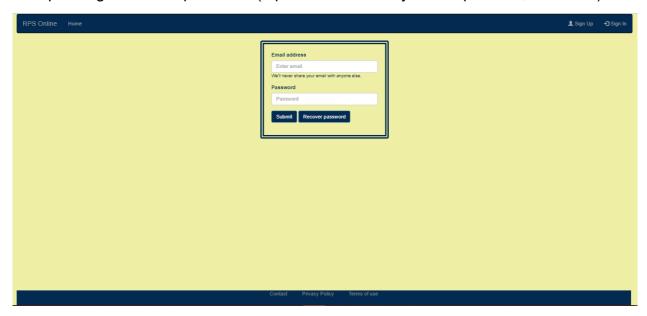
RPS Online is specifically designed to provide the user a set of tools to perform an integrated landmark-based resistant shape analysis of 2D and 3D datasets. All its functionalities can be invoked from the user interface, which is organized as a main page with two pop-ups, Sign-up and Sign-in:



The first time user in RPS Online has to Sign up to create a new account:



Next, the Sign in pop-up window enables to get inside RPS Online by entering the corresponding email and password (a password recovery is also possible, if needed):



When the user is finally inside RPS Online, the dashboard management view is displayed.

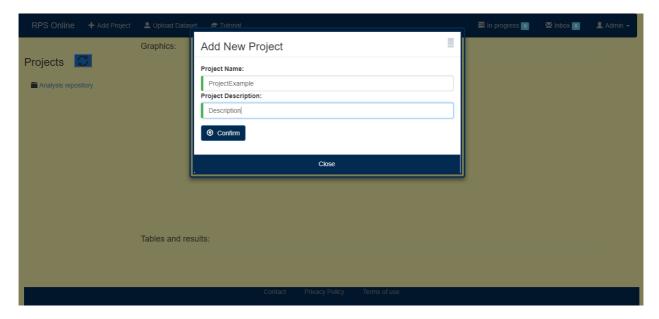
#### Dashboard area

RPS Online organizes datasets and analyses into projects, which are intended to be self-contained. The main area of the app thus offers a general overview of the project items: The Analysis Repository and the tree of projects, datasets and their analyses (left) and the Graphics and Table and Results tabs (center)

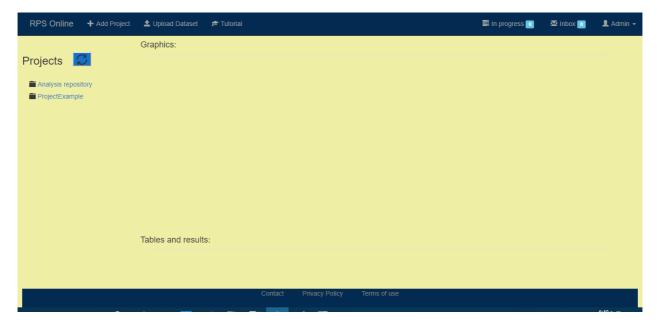


## **Creating a Project & Loading a New Dataset**

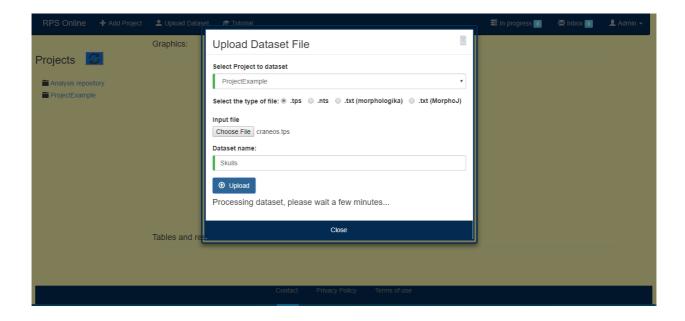
To create a new project, the command Add Project from the toolbar must be selected. A pop-up window will be displayed where the Project Name and the Project Description must be filled in:



Once this window is completed, the name of the new project will automatically be attached to the tree on the left:



If the user wants instead to add a new dataset to an existing project, the command Upload Dataset in the toolbar must be selected. After choosing the project, the name of the new dataset must be filled in:



The uploaded dataset will be automatically displayed in the Graphics tab, while the Cartesian coordinates of its objects will be given below in the Tables and Results tab. In addition, the new dataset will be automatically attached to the project in the left tree and all the objects included in it will be nested in a new level of the tree:



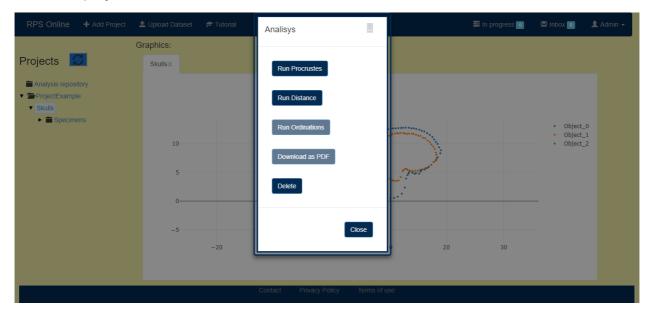
Note that when passing the mouse over any landmark in a graphical display, the corresponding Cartesian coordinates, landmark number and object number will be shown.

## MAIN MENU (by right-clicking on a file from the left tree)

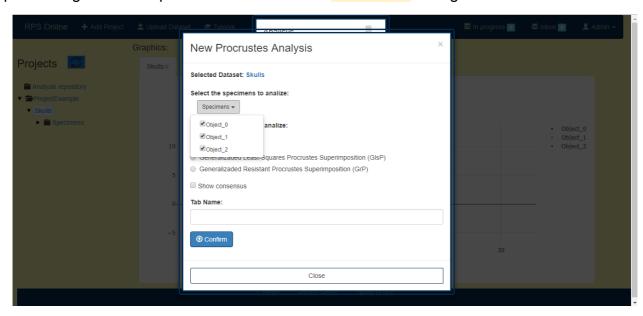
## **Run Procrustes Superimpositions**

The two available Procrustes superimpositions, GlsP and GrP, can be performed only on a dataset consisting of landmark configurations.

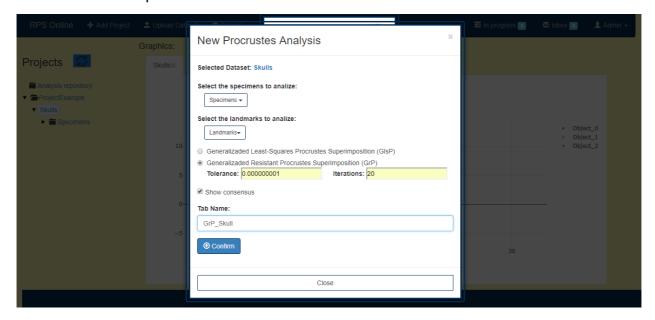
First, the user has to select the dataset to process by right-clicking on it from the tree on the left: e.g. the file Skulls (see capture). A pop-up menu highlighting all the available analyses for that type of data (superimpositions or distance matrices can be computed on a set of landmark configurations, while ordinations are based on a distance matrices) will be next displayed:



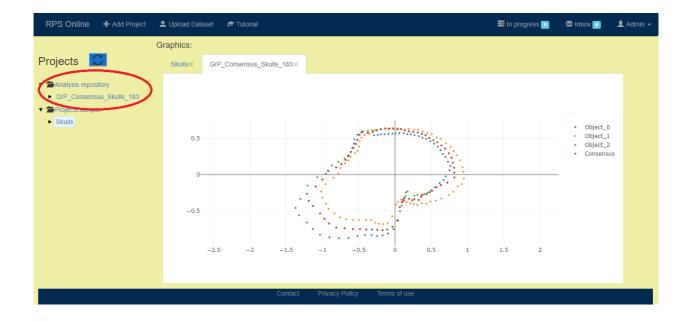
Once the desired type of analysis is selected, a pop-up window is displayed to fill-in the name of the output dataset. There, it is possible is to unselect some objects and/or landmarks (from every object) from the default to not be included in the subsequent processing. And it is optional to show the final Consensus configuration.



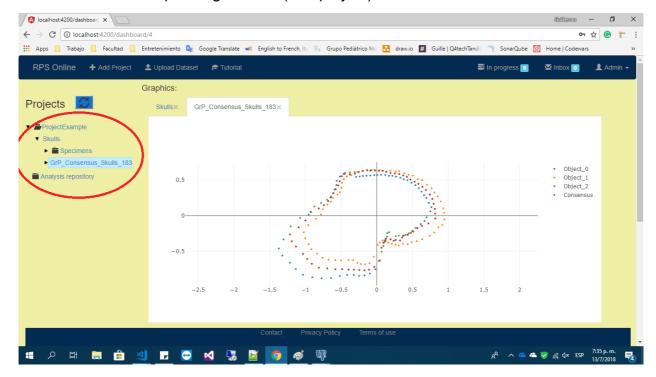
When the Generalized resistant Procrustes superimposition (GrP) is selected, the pop-up menu enables to specify: the desired Tolerance, the maximum number of Iterations and the name of the output dataset.



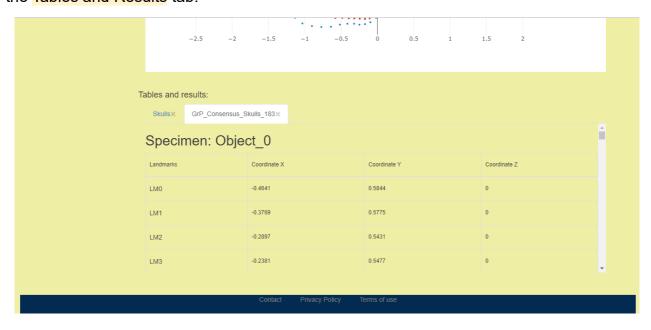
Once finished, the obtained superimposition is automatically added to the Analysis Repository and can be visualized in the Graphics tab:



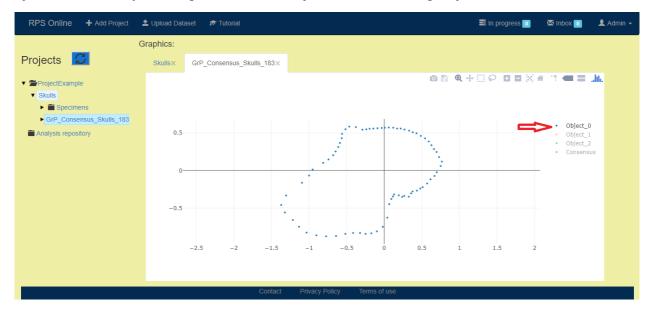
When the blue update button is clicked, each analysis in the Analysis Repository is attached to the corresponding dataset (and project) in the tree on the left.



The Cartesian coordinates of the visualized superimposed objects are displayed below, in the Tables and Results tab:

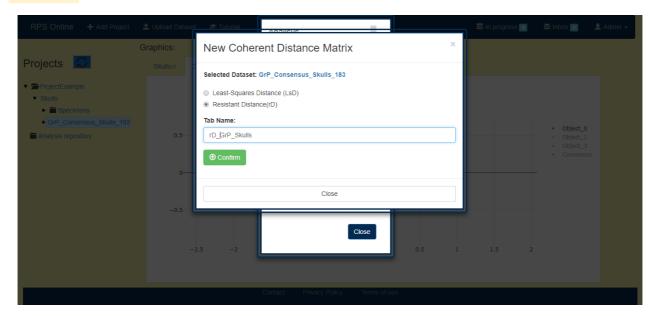


Note that in the **Graphics** tab it is possible to hide an object by clicking on it from the list of object's names. By clicking twice on an object, the remaining objects are instead hidden:

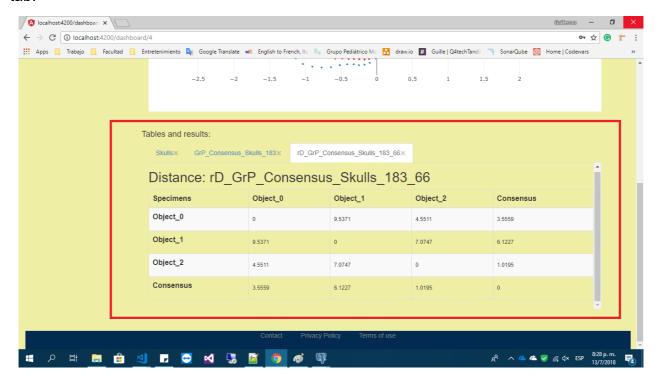


## **Compute Distance Matrix**

To compute a Distance Matrix between objects following a superimposition, the user has to select the corresponding superimposed data from the tree on the left and right-click on it: the pop-up menu of available analyses is displayed again and after selecting Run Distances the two choices are shown:

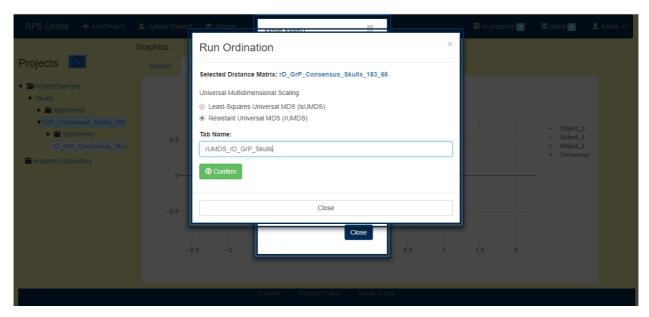


Once computed, the corresponding distance matrix is shown in the **Tables and Results** tab:

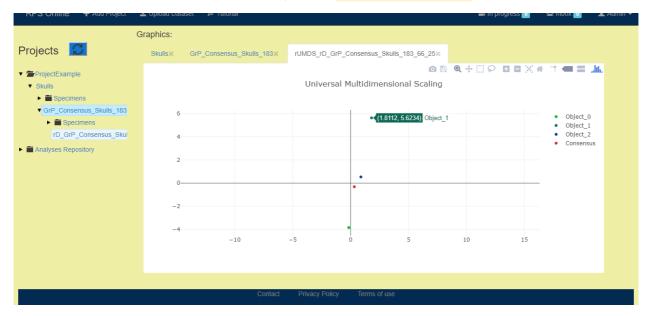


#### **Run Ordination**

Finally, when an ordination from a computed distance matrix is desired the user has to right-click on the corresponding distance matrix file from the tree. The pop-up menu offering both types of ordinations is then shown and the name of the output must be filled in:



Finally, the computed ordination is shown in the Graphics tab while the corresponding Cartesian coordinates are shown below, in the Tables and Results tab:



#### **Additional Comments**

The tree structure on the left reserves the first level for projects (e.g. ProjectExample); the second level for the dataset related to that project (e.g. Skulls) and the third level includes:

- the list of landmark configurations, under the tag Specimens;
- the list of analyses performed on each of them;
- eventually, it may include a Consensus configuration if the performed analysis was a superimposition.

Performed analyses on each file can be distinguished by the automatic chain of prefixes in their names (from right to left, as successive layers):

- GrP\_ (for a Generalized resistant Procrustes superimposition);
- GlsP (for a Generalized least squares Procrustes superimposition)
- rD\_ (for a resistant Distance)
- IsD (for a least squares Distance)
- rUMDS\_ (for a resistant UMDS)
- IsUMDS (for a least squares UMDS)
- 2D (for a two dimensional UMDS)
- 3D\_ (for a three dimensional UMDS)

All the performed analysis and computations can be downloaded by selecting the command Download as PDF when right-clicking on a specific file.