prepNFB User Manual 1.0

prepNFB

To conduct a NFB training session with OpenNFT, certain files must be prepared and made accessible to the software. These files include, but are not limited to, the subject-specific ROIs, an echo planar imaging (EPI) scan for real-time motion correction, a high-resolution anatomical scan to visualize whole-brain activity changes and a protocol file detailing the occurrences of each experimental condition during a training run (e.g, baseline, self-regulation, task). To house these files, directories specific to each participant must be created and correctly assigned in the OpenNFT GUI. This entire preparatory process can prove to be complex, particularly within the time sensitive constraints of the MRI environment. We therefore automated this process and standardized the preparatory workflow by developing the open-source toolbox *prepNFB* which is available on GitHub (https://github.com/lucp88/prepNFB). Written in MATLAB and comprising custom made functions as well as adapted SPM routines, *prep*NFB offers a comprehensive GUI-based interface that can be fully tailored to accommodate any OpenNFT-based neurofeedback experiment (**fig. 3**).

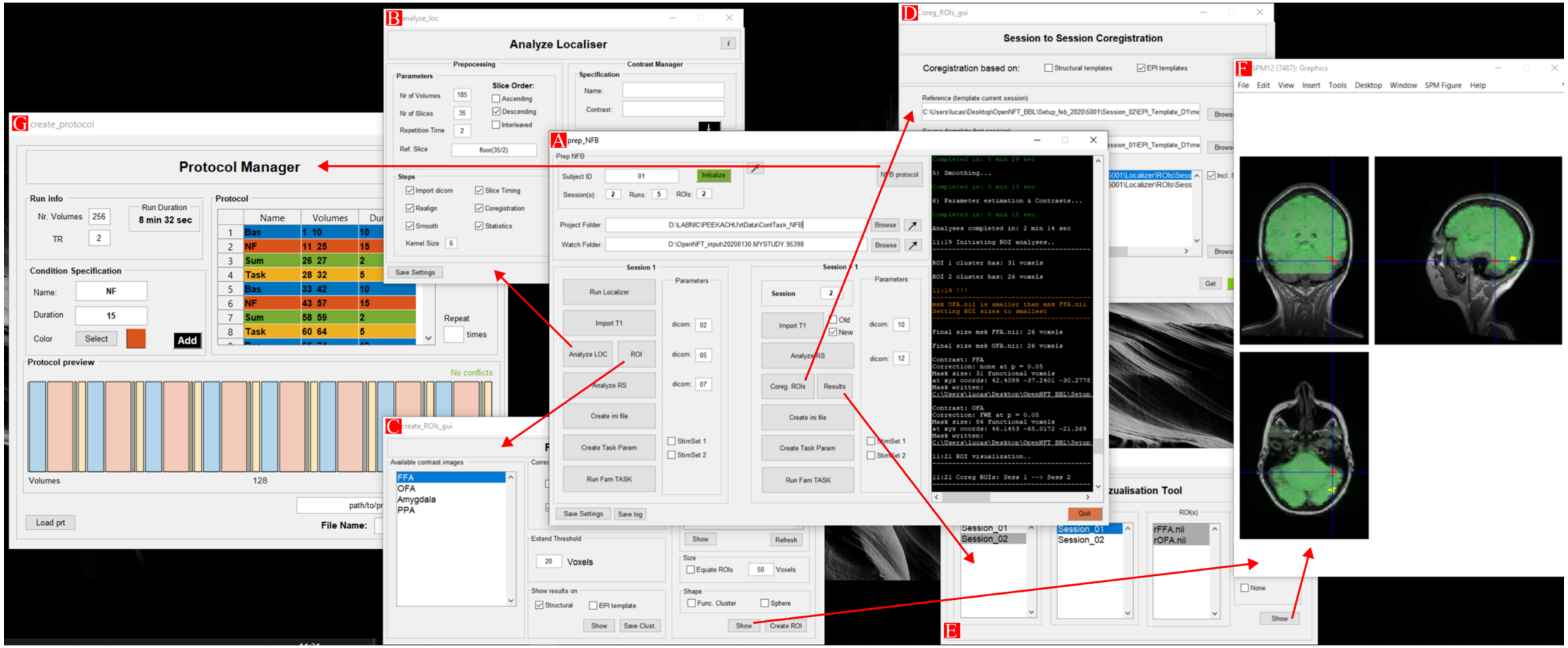

Figure 3. An expansive view of the prepNFB toolbox, featuring the primary graphical user interface **(A)** and a variety of essential tools available to the user that are launched by engaging the respective buttons. These tools, detailed further in the subsequent figures, include: **(B)** The Analyze Localizer tool (see Figure 4), **(C)** The ROI Delineation Tool (see Figure 5), **(D)** The Session-to-Session ROI Coregistration tool (see Figure 6), **(E)** The ROI Visualization Tool (also see Figure 7), **(F)** The interactive Whole Brain Results Window which is activated from both the ROI Delineation- and Visualization Tool (see Figure 8). **(G)** The OpenNFT Protocol Manager (see Figure 9).

Functionalities and Architecture

The main interface (**fig. 4**) is divided into four sections; a top panel **(A)**, bottom left **(B)** and bottom right panel **(C)** and a large status window **(D)** where relevant messages are displayed for the user. Conveniently, all settings can be saved at any moment for easy and automatic loading upon the next launch (bottom left).

Initialization of the NFB Training Session

Panel A (**fig. 4**) allows for the input of participant details such as *subject ID* number, the number of training *Sessions* and *Runs* to be conducted as well as the *'Project Folder'* **(A1)** and the *'Watch Folder'* **(A2)** directories. The '*Watch Folder'* is a key feature of prepNFB as it points to the directory where the MR images are written to in real time. This allows *prep*NFB to connect, import, and analyze the relevant brain data needed for the steps detailed below. The functional images are retrieved from the '*Watch Folder'* based on the DICOM series numbers specified in the *Parameters* panel. Once the participant details are submitted in panel **A**, the 'Initialize' feature (green button) is used to create an OpenNFT compatible directory tree on the real-time processing PC according to the ID number, sessions, runs and ROIs specifications. Panel **B** and **C** (**fig. 4**) provide access to a series of analytical tools. Specifically, panel **B** (**fig. 4**) presents all options pertaining to the first NFB training session while panel C includes features needed for subsequent training sessions (to be specified in the *Session* edit box).

First NFB Training Session

Key features for the first session panel (**fig. 4B**) include the *'Run Localizer'* (B1) and *'Analyze Localizer'* tools (**fig. 5**). These jointly facilitate the flexible execution and analysis (i.e., standard preprocessing and first-level statistical analysis) of a functional localizer task. The resulting statistical contrast images are automatically loaded into the *'ROI Delineation'* tool (**fig. 6**), which enables the visual identification and selection of one or more clusters of interest based on pre-specified statistical contrasts. These clusters can then be edited as needed and converted into binary mask files which are utilized by OpenNFT as ROIs for NFB (see **fig. 6** legend for details). The entire process, from executing the localizer task to delineating the ROIs, can be completed in under 10 minutes, which enables the preparation to be carried out directly before the first NFB training while the participant is already in the scanner, possibly during a pre-training resting state acquisition.

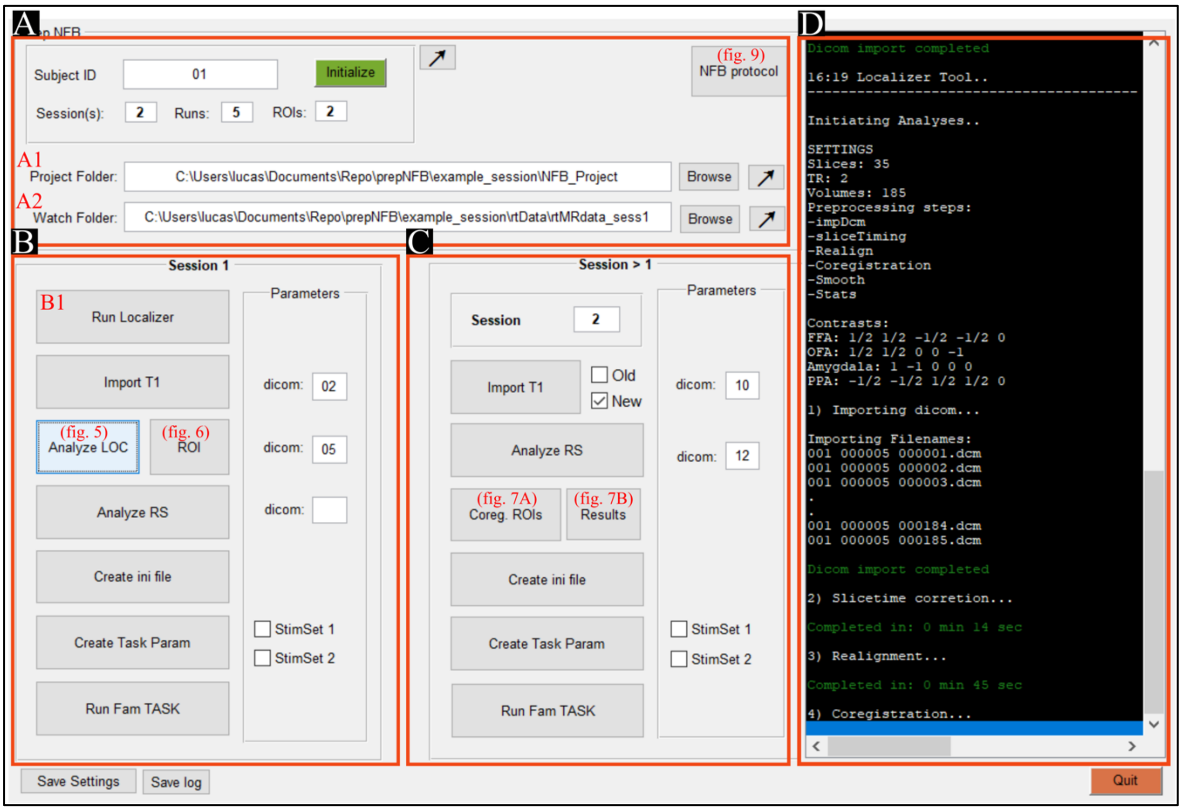

Figure 4 The main prepNFB interface is accessible through MATLAB's command line or a shortcut. The interface comprises four sections: **(A)** The participant details and session initialization panel, **(B)** panel with features pertaining to the first NFB training session, **(C)** the panel containing tools for subsequent training sessions, and **(D)** a large status window displaying user-relevant messages. In the current example prepNFB is analyzing functional localizer data. For more details, please refer to the main text.

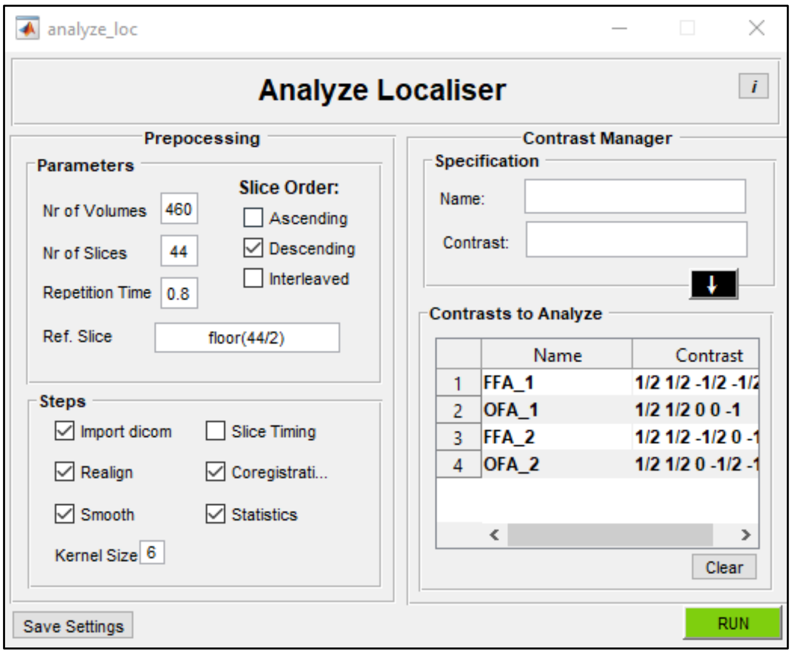

Figure 5 Analyze Localizer Tool. Following completion of a functional localizer task, this tool carries out standard preprocessing and first-level fMRI data analysis. The Parameters panel in the Preprocessing section allows users to specify the required settings. Within the Steps panel, users can either opt for an entire preprocessing routine, from Import dicom files to Statistics, or choose specific components if a partial routine is required. The Contrast Manager allows users to name and specify a statistical contrast according to SPM standards, which can then be added to the Contrasts to Analyze list using the black-arrow button. All settings can be conveniently saved for automatic loading upon subsequent usage. The Run button initiates the workflow.

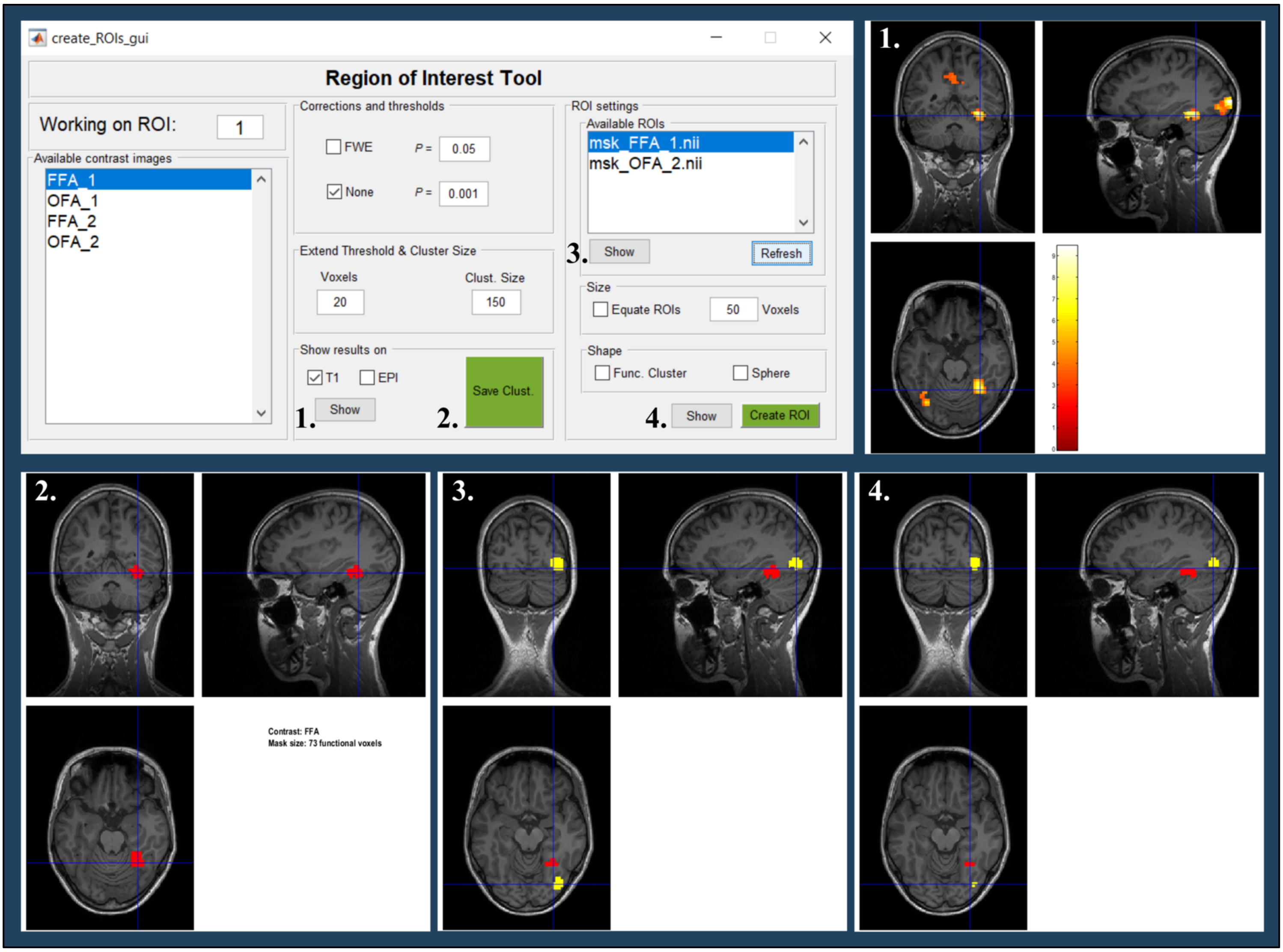

Figure 6. Region of Interest Tool. This feature enables an interactive and flexible delineation of ROIs using the contrast images produced by the Analyze Localizer Tool (figure 5). Users operate the tool column-wise, from left to right, first creating a larger ROI cluster from which the final voxels are selected. To start, the desired contrast image is chosen from the *Available Contrast Images* window (e.g., FFA_1), with *statistical threshold*, *multiple comparison correction*, and *extent threshold* defined in the center column (default values as shown). When clicking *Show*, the user's selected contrast image is displayed on the participant's Anatomical (T1) or EPI scan in a modified SPM12 orthogonal view window **(1)**. Users can navigate to the cluster of interest, select the peak voxel or manually position the cross-hair, and *Save Clust.* to generate an intermediary binarized ROI file **(2** – e.g., *msk_FFA_1.nii***)**. This file is defined by the final cross-hair position and its N neighboring cluster voxels, as defined in the *Clust. Size* box. If *N* exceeds the number of voxels in the cluster, the user is presented with the option to *trim to max* (take all available voxels) or *adjust* the cluster size. Intermediary cluster results immediately replace the whole-brain contrast results in the orthogonal view window **(1 → 2)**.

This process is repeated for each ROI. Upon generating all intermediary ROI files, users finalize the process in the last column: *ROI Settings* panel. *Refreshing* the *Available ROIs* section displays all generated ROI files. Users can select from this pool of available ROIs, choosing only those they wish to visualize (normally all). The selected ROIs can then be projected onto the participant's T1 image using the *Show* function **(3)**. If overlap or errors are detected, users can return to specific ROIs and modify parameters as needed (e.g., adjusting the statistical threshold). Once all intermediary ROI images are satisfactory, the final ROI files are created. To do so, users may opt to limit and *equate the ROI* sizes to a pre-specified number of voxels (*N*) in the *Size* panel. In this case only the *N* voxels from the intermediary cluster with the highest T-value for the ROI's contrast are retained. The final *Shape* panel offers users the ability to choose between two different final ROI delineation routines - one that is based on the shape of the functional cluster (*Func. Clust.*), and another that forms a *Sphere* composed of N voxels around the peak or crosshair voxel.

The *Show* function **(4)** executes the finalization routine, generating the final ROI files according to the specified settings. As part of this process, *prepNFB* automatically checks (and warns) for shared voxels between the ROIs and removes any overlap. The finalized ROIs are then displayed onto the same T1 image in the orthogonal view window for a final visual verification **(4.)**. Once the results are deemed satisfactory, the *Create ROI* function prompts users to name the final ROI files, which are automatically written to the appropriate OpenNFT directories. Additionally, a record of all actions, parameters settings and results is continuously updated and saved to a log.txt file for later reference.

To see a video example of the above, please go to: https://www.youtube.com/watch?v=bswgG1_mOtE&ab_channel=Luc

Subsequent NFB Training Sessions

To correct for inevitable variations in participant head positioning across different training sessions, **figure 4C** includes a critical feature for multi-session NFB studies; the *'Session to Session (ROI) Coregistration'* tool **(fig. 7A).** As the name implies, this routine facilitates the coregistration of the ROIs delineated in the first session across all subsequent training sessions, ensuring that feedback is consistently drawn from the same anatomical regions despite shifts in head positioning. This is achieved by acquiring a new structural (T1) or functional (EPI) scan for each training session, and coregistering the first session's scan of the same modality and the original ROIs to this newly acquired image. This approach effectively aligns the originally delineated ROIs with the current head position in the scanner. After the coregistration step is completed, the *Results* function (**fig. 4C**) opens a *ROI Visualization* tool **(fig. 7B)** that allows for the visualization of the results onto relevant EPI and/or structural images for visual quality assessment **(fig. 7C)**. These positions can then be directly compared to the original as delineated in the first session (not shown).

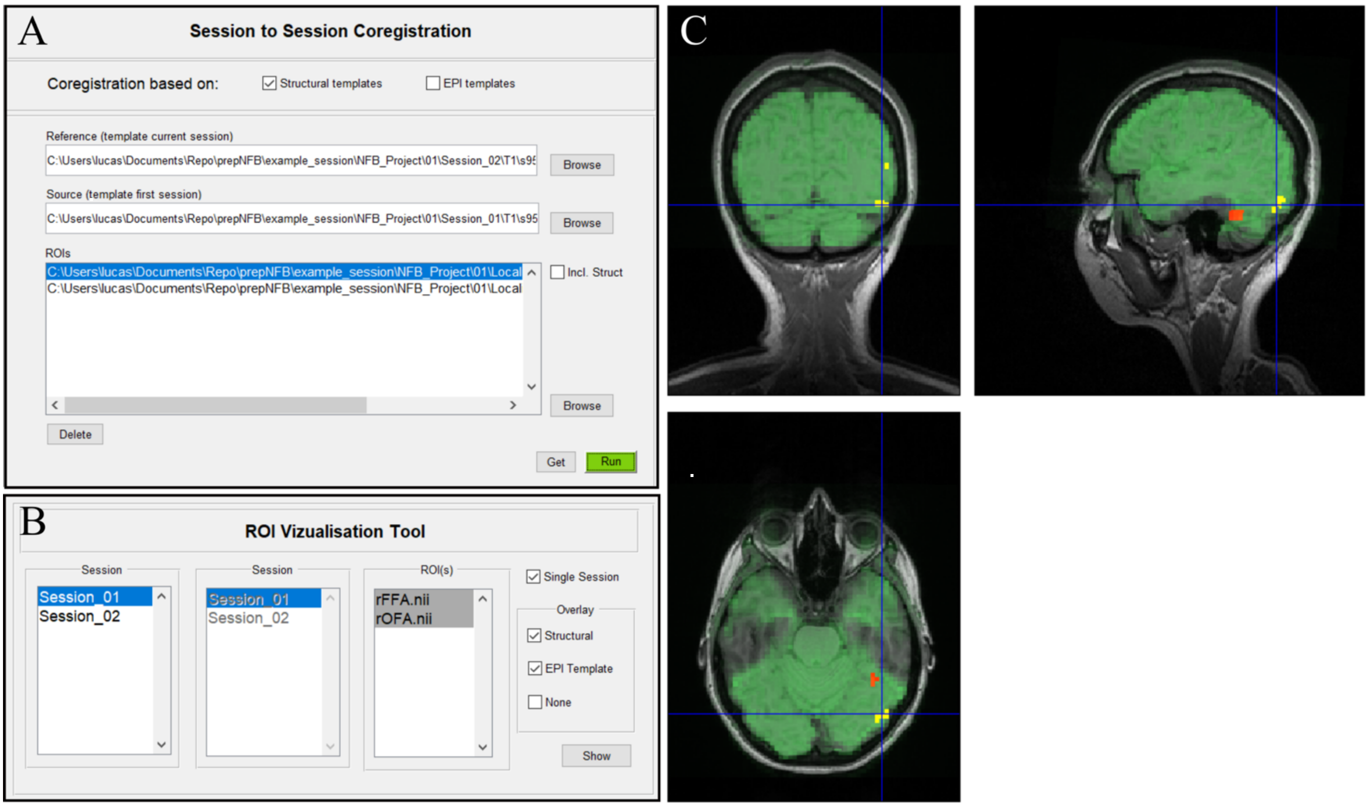

Figure 7. Session to Session Coregistration Tool. This functionality coregisters the original ROI file(s), as defined during the delineation process (figure 6), to new head positions for the same participant during subsequent NFB training sessions.

(A) In the *Session-to-Session Coregistration* window, users first specify whether coregistration will be based on *Structural* (T1) or functional (*EPI*) templates. They then have the option to either manually *Browse* for the *Reference*, *Source*, and *ROI* images or to use the *Get* function (bottom right), which automatically locates and loads the images based on information provided in the main interface. For EPI-based coregistration in the absence of a new T1 image acquisition, users can choose to include the original T1 image from the ROI delineation session in the coregistration process by selecting the *Incl. Struct.* option. Upon clicking *Run*, the selected information and parameters are passed to an adapted version of the SPM12 coregistration function for execution.

(B) Following coregistration, the *ROI Visualization Tool* facilitates visual inspection for proper alignment of the coregistered ROIs with the present head position. Users have the option to project one or more ROIs from a *Single Session* **(C)** or juxtapose two sessions side by side for comparison. All available sessions are automatically listed in the *Session* panels. Users can choose to *Show* the ROIs onto a structural and/or EPI template. **(C)** When both structural and EPI are selected as *overlays*, the ROIs (solid colors) and the current session's EPI data (green overlay) are mapped onto the participant's T1 scan.

Lastly, the main user interface (**fig. 4A**) includes a tool named 'Protocol Manager' **(fig. 8)**. This tool is specifically engineered to generate, review, and save new experimental protocol files compatible with OpenNFT (.json), as well as to load and modify existing protocols. Conveniently, the tool automatically checks for potential conflicts in single volumes being assigned to multiple conditions like regulation *and* baseline and offers an automatic 'resolve' option that can be engaged to resolve detected conflicts.

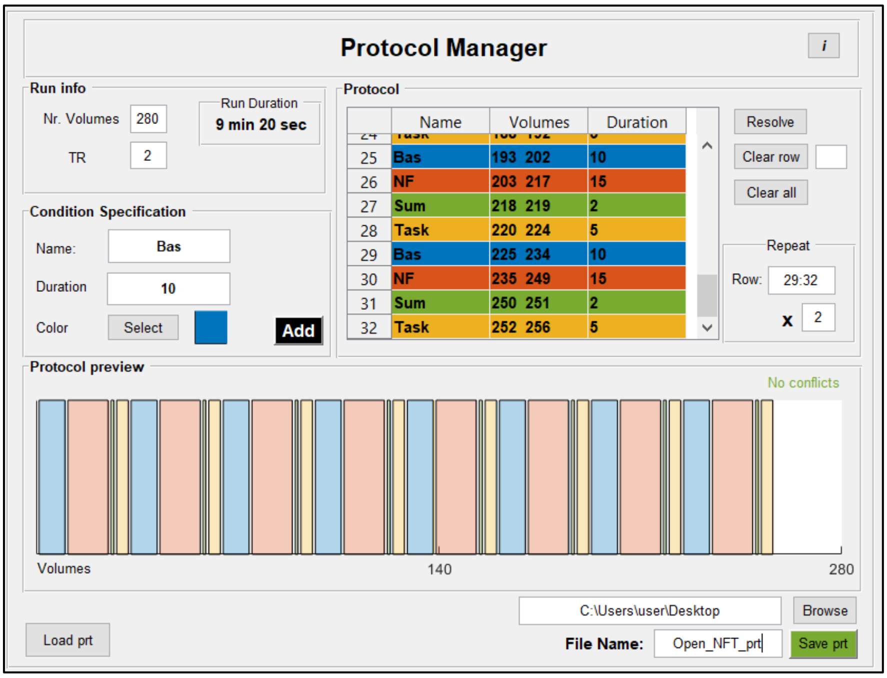

Figure 8. The Protocol Manager. This tool enables users to create a NFB protocol file that is used by the OpenNFT toolbox to execute the different conditions (e.g., baseline, regulation, task). In the *Run Info* panel users provide the number of volumes per run and the repetition time (TR) which is needed to allocate the correct number of volumes to each condition. Subsequently, the first sequence of conditions is specified (e.g., Bas,10 volumes, color: Blue | NF, 15 volumes, color: Red | Sum, 2 volumes, color green | etc.). Each time a condition is added (black button) it appears in the Protocol table and the Protocol preview panel. Overlap is automatically detected and displayed as overlapping panes in the preview panel. The Resolve function is used to automatically correct such conflicts. Once the initial sequence is specified, users can Repeat Row N:N for an x number of times, which automatically extends the protocol. The Run Duration is updated dynamically according to the data in the Protocol table. Prior to saving the protocol with Save prt, users can define a directory and a File Name.