Functional MRI Statistical Software Packages: A Comparative Analysis

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Abstract: Currently, there are many choices of software packages for the analysis of fMRI data, each offering many options. Since no one package can meet the needs of all fMRI laboratories, it is helpful to know what each package offers. Several software programs were evaluated for comparison of their documentation, ease of learning and use, referencing, data input steps required, types of statistical methods offered, and output choices. The functionality of each package was detailed and discussed. AFNI 2.01, SPM96, Stimulate 5.0, MEDIMAX 2.01, and FIT were tested. FIASCO, Yale, and MEDx 2.0 were described but not tested. A description of each package is provided. *Hum. Brain Mapping 6:73–84, 1998.* © 1998 Wiley-Liss, Inc.

Key words: functional imaging; fMRI; data analysis; brain

INTRODUCTION

Functional MRI (fMRI) is evolving at a fast pace as a neuroimaging research tool. This rapidly changing research has benefited from many of the lessons learned from PET. Since fMRI is noninvasive, less expensive, and more accessible than PET, many research centers are currently setting up fMRI facilities. Among the initial dilemmas for new imaging researchers is to determine how best to design and conduct studies, and to analyze and draw conclusions from the data collected.

There are many issues to be considered in terms of obtaining meaningful and generalizable study results. Some of these issues in fMRI cognitive activation studies include the determination of an appropriate sample size, the number and duration of cognitive tasks, the significance threshold, and the appropriate test statistic. Many of these methodologic concerns have been addressed in the PET literature [e.g., Poline and Mazoyer, 1993; McColl et al., 1994; Kapur et al., 1995; Arndt et al., 1995, 1996; Andreasen et al., 1996; Worsley, 1995, 1996; Gullion et al., 1996; McIntosh et al., 1996] and in the fMRI literature [e.g., Bandettini et al., 1993; Forman et al., 1995]. These types of decisions are often made in the context of the particular research question to be studied, and in terms of the existing knowledge about the phenomenon of interest, as well as the research and data analysis tools to be used.

Extensive data manipulation is necessary to draw conclusions from fMRI data, and since there are many

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available options for data analysis, a new fMRI research group will need to consider, ''Which available software package will best meet our needs?'' This question is the topic of this paper. The software packages are not interchangeable; in fact, many contain different procedures for the same data-processing steps. Each choice may affect all subsequent analyses. Furthermore, no one package is comprehensive.

Some laboratories have created their own software. This approach has advantages and disadvantages. One problem when comparing findings across laboratories is that more variability is introduced into the literature as more packages are used to analyze the data. Also, it is possible that individual packages could have mistakes, since thorough debugging can only be achieved after extensive use. And because of the complexities involved, it is also difficult for a researcher to fully understand a particular package unless the individual helped to design it.

The aim of this paper was to examine the available fMRI software packages and to compare the features each offers. It is not the intent of this paper to discuss the statistical methodologies or to endorse one method over another. Rather, it is to serve as an introductory aid to choosing a package for analysis. We delineate and comment on many factors such as: ease in learning and using the software, referencing, data input steps required, types of statistical methods offered, and display of the functional image. Functions and data transformations are detailed for each package.

METHODS

Locating fMRI software packages

Software packages were identified through examination of published fMRI studies, reference sections, and use of Internet search engines (e.g., Alta Vista, Yahoo, or WebCrawler) to conduct exploratory searches. Key words (e.g., fMRI analysis software packages) were identified to limit the search to fMRI or to imaging software. Functional MRI researchers were also consulted to identify fMRI data analysis software. We identified several packages and were able to obtain the individual Internet addresses (Universal Resource Locator; URL) for each package.

Inclusion/exclusion criteria

We include a description of all freeware, shareware, and commercial packages that were identified through our searches. These are: AFNI 2.01 (Analysis of Functional NeuroImages) [Cox, 1996], SPM96 (Statistical Parametric Mapping) [Friston et al., 1991], STIMULATE 5.0 [Strupp, 1996], Yale [Skudlarski et al., 1995], MEDIMAX 2.01 [Infographics Group, 1995], FIASCO (Functional Imaging Analysis Software-Computational Olio) [Eddy et al., 1996a], MEDx 2.0 [Sensor Systems Inc., 1996], and FIT (Functional Imaging Toolkit) [Arnholt, 1997].

For testing, the package had to be freeware and compatible, or easily converted to a Silicon Graphics (SG) platform. An exception to this was FIT, which was tested on an Intel platform. The software also needed to be able to read GE or ANALYZE [Robb and Hanson, 1991] format image data. ANALYZE is a commercially available software program. When a package did not meet these requirements, a partial evaluation was performed based on the program's manual, published documentation, and communications with the original authors.

Features examined

The features described below were included in our evaluation of each software package. Some of these items were rated on a 5-point difficulty scale, where 1 was the most difficult and 5 was very easy. Ratings were given only for features tested. The functionality of each package in terms of data transformations and functions performed was also reviewed. An attempt was made in this report to include citations for all relevant peer-reviewed journal articles that described functional components of the packages. Some packages provided algorithms and formulas in appendices of the user's manual.

Address/availability

The URL for each software package was noted, including information regarding whether the package is commercially available, freeware (free to users), or shareware (available to users with a request to donate money to the authors to help offset expenses).

Platform/operating system

The software and hardware requirements for each package were included in our survey. The base language of the program and the source code availability were documented.

Documentation

Packages were rated on the completeness of written instructions for installation, including downloading, compiling, and setup. The ease of learning and operating each package was rated. The inclusion / exclusion of a graphical user interface (GUI) was listed for each package. The GUI is a feature that allows software users to point and click on an item to invoke routines.

Referencing

The availability and source for instructions for program use, formulas, and descriptions of routines were included for each package. The year each package was first released into the public domain was noted as an index of the program's maturity and stability.

Preprocessing steps

The preprocessing steps necessary to convert fMRI data from native format to a form recognized by each software package were evaluated. Our image data were collected using the BOLD (blood oxygen level-dependent) method from a GE 1.5T Signa Scanner (Milwaukee, WI) retrofitted with Echo-planar imaging (EPI) by Advanced NMR Systems (Wilmington, MA).

Image realignment

Inclusion / exclusion of this feature was documented in our evaluation.

Input of data

The capability of each software package to input 2D, 3D, or 4D (i.e., the addition of the time dimension) image data was examined.

Types of reference (input) functions

Functional MRI experimental design typically takes the form of presentation of stimuli or task conditions in alternation. A waveform is chosen that closely models the time series of the task conditions. The relationship between the reference waveform and the fMRI time series data is assessed to determine which brain pixels are activated in a similar pattern with the reference function. The flexibility and availability of reference functions (e.g., sine wave, square wave, use of lag time, and best fit) with each package were included in the evaluation.

Statistical analyses

The statistical analyses available and other options, such as shielding some of the data from analysis, were evaluated for each package.

Image display

Image display features were evaluated for each package, including image manipulation tools, output options (e.g., regions, locations, and size), summary statistics, post hoc analyses, and whether the image could be viewed in three orthogonal planes.

Region of interest (ROI) analyses

The availability of ROI routines was documented.

Spatial transformation

Spatial transformations are important preprocessing steps in the analysis of neuroimaging data. These steps include realignment, normalization, and smoothing. Image realignment of a time series of scans attempts to minimize the effects of subject head movement. Since the BOLD effects that we are interested in are small, and the statistical analyses are based on voxel level intensity changes over time, even small object head movement can confound data analysis. Additionally, head movement during scanning can occur from multiple sources. Intersubject comparisons require the images to be spatially normalized to a standard brain. Spatial smoothing helps to reduce intersubject differences in anatomy and also increases the signal-to-noise ratio. The optimal width of the smoothing filter is chosen to closely match the size of the region which is activated [Worsley and Friston, 1995].

Statistical model

The reliance on parametric vs. nonparametric statistics was documented. For the packages that relied on parametric statistics, the method of accounting for autocorrelations in the data was noted. A high-pass filter is often used to remove cardiac and respiratory signals and other low-frequency drifts in the data that add to error and can correlate with the reference function, confounding task-related neural activations.

Correction for multiple comparisons

An enormous number ($\approx 64^2$) of voxel-level analyses are run on the data, inflating the type 1 error rate. Therefore, a correction for multiple comparisons is necessary. The type of correction each package utilized was assessed.

RESULTS

Table I presents an evaluation of the available features for each software package included in this study. Eight packages in all were examined. Five met inclusion criteria for testing, while three (MEDx, FIASCO, and Yale) were only partially evaluated. MEDx was excluded from testing because it is commercial software. FIASCO was not ready for distribution at the time of the evaluation. However, several laboratories within the University of Pittsburgh are using this software [W. Eddy, personal communication]. The Yale package was developed for specific computer workstations at Yale University. According to the author, there are currently no groups outside of Yale University using this package [P. Skudlarski, personal communication]. Each routine within this package may be viewed at the Internet site and downloaded individually. However, many of the scripts must be rewritten and individualized to the user's computer. A detailed list of the functions and data transformations for each package is presented in Table II.

Platform/operating system

While all of the packages support multiple platforms, AFNI and MEDx seem to be the most versatile. SPM and the Yale package require MATLAB [Mathworks, 1997], whereas AFNI, Stimulate, MEDx, MEDIMAX, FIASCO, and FIT do not. MATLAB is a high-level commercial programming package. Most of the other programs were written in C and provide source code.

Documentation

AFNI, SPM, Stimulate, MEDIMAX, and FIT provide instructions for downloading, compiling, and setting up the software. FIT uses an installation routine which automatically copies the executable version to a specified directory. We were able to set up AFNI, SPM, Stimulate, and FIT without difficulty. AFNI provided an ancillary image utility toolkit, Netpbm, which is also available from the Internet. We found the instructions for setting up MEDIMAX somewhat confusing, but we were able to successfully install the software. MEDIMAX lists the names of four ancillary utility packages, but the user must obtain these independently (two of these files are part of Netpbm). AFNI, Stimulate, Yale, MEDx, FIT, and MEDIMAX provide a user's manual. A user's manual for FIASCO is currently being written according to the authors [W. Eddy, personal communication]. SPM does not provide a formal user's manual. However, the authors monitor an E-mail discussion group. An SPM course is offered annually for a fee, although the course notes can be downloaded from their Web site. The 1997 course notes include illustrated guidelines for the use of SPM96. Most of the other packages provide E-mail addresses to document bugs or make requests for future upgrades. An annual workshop to learn Stimulate is offered for a fee. MEDx is the only package that provides full technical support.

AFNI, SPM, Stimulate, FIT, and MEDx are GUI-based, whereas FIASCO and Yale have a modular organization. The modular structure is hierarchical. Each routine invokes the next routine, so that users can execute a single command to run an entire series of data steps. AFNI contains a supplemental manual for C-literate users who would like to write external programs that may be run with AFNI. AFNI, SPM, MEDx, and FIT have GUI-based on-line help screens. The help screens provide definitions and detailed explanations about particular functions. SPM provides selected program manual pages through the on-line help system.

In our ratings, we differentiated between learning to use (including understanding how the routines are performed) and operating the packages. We found that all of the packages tested were easy to operate once we were comfortable with the package; however, the ease in understanding how the routines are performed as well as learning to use each feature varied between packages. The ease of operation was primarily due to the GUI-based nature of these packages, and to the defaults and/or examples that were often provided. We found FIT to be the easiest package to learn because the software has an interface and performance similar to those of other Windows-based software. AFNI and Stimulate both had an appealing and similar layout. We found navigating through the packages extremely easy, but both took some time to reach a level of proficiency. Stimulate received a slightly lower rating than AFNI for ease of learning because AFNI's features are more polished. We rated SPM as more difficult to learn because it took concerted effort to identify and understand how each routine operated. This may be due in part to the fact that SPM was initially developed for PET, with fMRI routines added to more recent versions.

Referencing

AFNI [Cox, 1996], SPM [Friston et al., 1991], and FIASCO [Eddy et al., 1996a] are described in peer-reviewed journal articles written by the authors of the

TABLE I. Features included in each fMRI analysis software package

Features	AFNI 2.01, analysis of functional neuroImages	SPM96, statistical parametric mapping	Stimulate 5.0
1) Address/availability	http://www.biophysics.mcw.edu Freeware	http://www.fil.ion.bpmf.ac.uk/spm Freeware	http://www.cmrr.drad.umn.edu Freeware
2) Platform/operating system	SGI, Sun Solaris, Linux, HP/Unix, 64 Mb	SGI, Sun DEC-Alpha/Unix, 128-Mb 300-Mb swap	Sun, Solaris, SGI/Unix, 128 Mb 1G swap
a) Base language/software	ANSI C, motif 1.2	MATLAB	C
b) Source code availability	Yes	Yes	No
c) Graphical display minimum	8-bit	No minimum	8-bit
3) Documentation/support	User's manual/on-line help	Course/course notes/on-line help, limited user's manual	User's manual/annual workshop
a) Ease of installation[a]	3	4	4
b) Ease of learning	4	2 (E-mail discussion list)	3
c) Ease of use	4	4	4
d) Graphical user interface	Yes	Yes	Yes
4) Referencing	User's manual; journal articles	Journal articles, book chapters, course notes	User's manual, workshop
a) First release	1995	PET (1990); fMRI (1994)	1995
5) Preprocessing steps	Raw images must be converted	Raw images converted to ANALYZE format	3 choices for image conversion: FID, Sdt, raw (all menu-driven)
6) Image realignment	2D in-slice only	Yes	No
7) Data input 2D, 3D, or 4D	2D, 3D, 4D	2D, 3D, 4D	2D, 3D, 4D
8) Types of reference functions	Image-based, multiple time-lagged, user-defined	Boxcar, half sine, 2 basis functions (modulated sine)	Boxcar, multiple time-lagged, user-defined
9) Statistical analyses	Correlation, t-test, ANOVA, descriptives	t-test, F-test, eigenimage analysis	t-test, correlation, temporal statistics
10) Display of image			
a) Image manipulation tools	Yes, various maps, modify palette, and threshold	Yes, image subtraction and addition	Many options, modify palette and threshold
b) Output of regions, locations, size	Yes	Yes	Yes
c) 3 orthogonal views	Yes	Yes	Yes
d) Statistical output	Correlation coefficients, probabilities, various maps Descriptive statistics, t-, F-statistics, *P*	z-, t-, and F-statistics, maps, *P*	Correlation map, time shift, % intensity change, descriptive statistics
11) ROI analyses	No	No	Yes

Features	MEDx 2.0	Yale package	FIASCO, functional imaging analysis software-computational olio
1) Address/availability	http://www.sensor.com Commercial package	http://www.mri.med.yale.edu/individual/pawel/fMRI3 Freeware	To be released Freeware

TABLE I. (continued)

Features	MEDx 2.0	Yale package	FIASCO, functional imaging analysis software-computational olio
2) Platform / operating system	HP, SGI, Linux, DEC Alpha, Solaris, SunOS / Unix	Sun, SGI / Matlab, 60 Mb	HP, SGI, DEC / Unix
a) Base language / software	C (computationally intensive parts) and Tcl	MATLAB	C
b) Source code availability	Tcl source is provided	Yes	Yes
c) Graphical display minimum	No minimum	No minimum	No minimum
3) Documentation / support	Extensive manual / tutorial / on-line help / technical support	User's manual	User's manual (currently not available)
a) Ease of installation[a]	N / A	2 (package was not made portable, requires rewriting)	N / A
b) Ease of learning	N / A	N / A	N / A
c) Ease of use	N / A	N / A	N / A
d) Graphical user interface	Yes	Yes	Package has heirarchical structure (main script invokes subsequent scripts, defaults available)
4) Referencing	User's manual, journal articles	User's manual	Journal articles
a) First release	1994 (NIH Intramural Research Program), 1996 (public domain)	1996	1995
5) Preprocessing steps	20 native file formats may be directly read in	Raw images must be converted	Raw images must be converted
6) Image realignment	Yes, AIR	2D in-slice only	Yes (in Fourier domain)
7) Data input 2D, 3D, 4D	2D	2D, 3D, 4D	
8) Types of reference functions	User-defined and loaded from ASCII files	Lag time, boxcar, user-defined	Multiparameter spline
9) Statistical analyses	t-test, ANOVA, Kolmogorov-Smirnov	t-test, % difference, power of Fourier components, correlation, Mann-Whitney	t-test, S-Plus software
10) Display of image			
a) Image manipulation tools	Yes	Yes	No
b) Output of regions, locations, size	Yes	Yes	No
c) 3 orthogonal views	Yes	No	No
d) Statistical output	Yes	Yes	Yes
11) ROI analyses	Yes	Yes	No

Features	MEDIMAX 2.01	FIT, functional imaging toolkit
1) Address / availability	http: / / www.alsace.u-strasbg.fr / ipb / medimax01 Freeware	http: / / www.concentric.net / ~arnholt / inidex.html

TABLE I. (continued)

Features	MEDIMAX 2.01	FIT, functional imaging toolkit
2) Platform / operating system	HP-UX, SGI / Unix, Linux, 64 Mb	Intel, Cyrix, AMD / Windows 95 / NT, 32Mb
a) Base language / software	C	C++
b) Source code availability	Yes	Yes
c) Graphical display minimum	8-bit	8-bit
3) Documentation / support	User's manual / on-line help	User's manual / on-line help
a) Ease of installation[a]	4	5
b) Ease of learning	2	5
c) Ease of use	3	4
d) Graphical user interface	Yes	Yes
4) Referencing	None	Unpublished doctoral thesis
a) First release	1996	1997
5) Preprocessing steps	Raw images must be converted	Raw images converted to TIFF format
6) Image realignment	No (scheduled for next version)	Yes (AIR)
7) Data input 2D, 3D, 4D	2D, 3D	2D, 3D
8) Types of reference functions	Image-based, boxcar, multiple time-lagged	Boxcar, sine, multiple time-lagged, slope
9) Statistical analyses	Correlation, ratio, change scores	Correlation, Kolmogorov-Smirnov, t-test, chi-square
10) Display of image		
a) Image manipulation tools	Change palette and threshold	Yes
b) Output of regions, locations, size	Only when ROIs are traced	Regions
c) 3 orthogonal views	No (scheduled for next version)	No
d) Statistical output	Yes	Correlation coefficients, probabilities, t-statistics
11) ROI analyses	Yes	Yes

[a]Numeric ratings were on a scale of 1–5; 1 indicated difficulty with the feature, and 5 indicated ease with the feature. N / A, not applicable; feature was not tested.

software. AFNI and SPM provide some of the algorithms used to perform analytic functions in the journal articles. The AFNI user's manual and the SPM on-line help system also contain the algorithms for many of the routines.

Preprocessing steps

MEDx, which supports 20 image formats, can read GE fMRI data, whereas the other packages require the images to be reformatted. SPM requires that images be converted to ANALYZE format to be read by the program. We were able to write a script that converts images to the same format as ANALYZE. The ANALYZE-formatted images were also used for Stimulate. Stimulate provides three menu-driven options to load images, and the user need only fill in the parameter values. To load an image to be read by AFNI, an external ".BRIK" file must be created which contains the image parameters. MEDIMAX users may convert images into BRUKER or Interfile formats or create GIF, PIC, PPM, or TIF images from raw images. Yale, FIASCO, and FIT also require images to be converted before being read into the program. SPM, AFNI, Stimulate, and Yale have routines to transform images to Talairach-Tournoux coordinate space [Talairach and Tournoux, 1988] for comparison of image data across subjects.

Types of reference (input) functions

Many of the packages contain options for reference functions (see Table I). Some of the packages (e.g., MEDx and AFNI) contain descriptions for users to create and store their own waveforms. Otherwise, the user must create and store waveform options (e.g., ASCII files) to be read by a program. AFNI, SPM, and Stimulate output the optimal lag. AFNI and Stimulate allow the user to select image-based reference functions. Multiple time-lagged waveforms may be created directly from the image, and these can be smoothed.

TABLE II. Functions and data transformations available in each software package

Features	AFNI 2.01	SPM96
1) Spatial transformation		
a) Realignment	Least-squares analysis	Least-squares analysis / multilinear regression
b) Normalization	3D piecewise affine	3D affine and nonlinear
c) Resampling	Nearest-neighbor, linear, cubic interpolation	Nearest-neighbor, trilinear, sinc interpolation
d) Smoothing	Convolution with Gaussian kernel and other filtering options	Convolution with Gaussian kernel
2) Statistical model	Parametric and nonparametric	Parametric (general linear model; GLM)
a) High-pass filter	Yes	Yes Temporal convolution
b) Accommodation of autocorrelations	Reduction in degrees of freedom	Extended GLM
3) Correlation for multiple comparisons	Bonferroni correction, Monte Carlo simulation	Gaussian random field theory, Euler characteristic

Features	Stimulate 5.0	MEDx 2.0	Yale package
1) Spatial transformation			
a) Realignment	Not available	Ratio (AIR)	Least-squares analysis
b) Normalization	3D affine	3D linear piecewise or linear affine	3D affine
c) Resampling	Nearest-neighbor, linear interpolstion	Nearest-neighbor, linear, sinc interpolation	Linear interpolation
d) Smoothing	Convolution with Gaussian kernel (via equivalent multiplication in Fourier space)	Convolution with Gaussian kernel and other filtering options	Convolution with Gaussian kernel median filtering
2) Statistical model	Parametric	Parametric and nonparametric	Parametric and nonparametric
a) High-pass filter	Yes (low- and band-pass available)	Yes, other filters Temporal convolution	Yes
b) Accommodation of autocorrelations	Not available	Available when SPM is used for statistical analysis	Not available
3) Correlation for multiple comparisons	Bonferroni correction via auxiliary package, http://www.cmrr.drad.umn.edu/software/cluster_prob.html	Bonferroni correction, Gaussian random field theory, Euler characteristic	Not available

Features	FIASCO	MEDIMAX 2.01	FIT
1) Spatial transformation			
a) Realignment	Least-squares analysis	Iterative optimization technique	Ratio (AIR)
b) Normalization	Not available	Not available	Not available
c) Resampling	Fourier interpolation	Nearest-neighbor, linear, sinc interpolation	Linear, sinc interpolation (AIR)
d) Smoothing	Not available	Convolution with Gaussian kernel and other filtering options	Filtering options
2) Statistical model	Nonparametric	Nonparametric	Parametric and nonparametric
a) High-pass filter	Not available	Yes	No, Laplacian, Sobel, median, and other filtering
b) Accommodation of autocorrelations	Reduction in degrees of freedom	Not available	Not available
3) Correlation for multiple comparisons	Cluster size threshold	Not available	Not available

Statistical analyses

Each package provides options for statistical tests. Some packages will also calculate and display descriptive statistics. The packages vary somewhat in the available options for statistical tests. All of the packages allow data to be shielded from analyses. For instance, the first task cycle or other collected data may be discarded or kept out of statistical analyses. Table I lists the options for each package.

Image display

SPM, Stimulate, AFNI, and MEDx allow data to be viewed in the three orthogonal views. AFNI has a 3D cluster routine that will examine a data set for clusters of a thresholded size and volume, and create an image map. This feature will output volumes, center of mass, and the highest voxel value. SPM, MEDx, Stimulate, and FIT also have cluster-detection routines. Stimulate, MEDx, and FIT have a movie tool that can be used to display a series of images to observe physiologic or subject motion. FIASCO is the only package that does not currently contain tools for image manipulation.

ROI analyses

MEDx, Yale, MEDIMAX, Stimulate, and FIT contain ROI routines. Capabilities vary among the packages.

Spatial transformation

Image realignment is necessary to correct for motion caused during acquisition of the studies. Sources of motion include head movements and environmental and physiological rhythms such as cardiac and respiratory cycles. MEDx and FIT perform image realignment using the AIR algorithm [Woods et al., 1992]. AIR sequentially aligns each volume to a reference volume, using a method which correlates voxel intensities. The images can then be resliced onto a new grid using linear or sinc interpolation. SPM uses a least-squares minimization of the difference (rather than ratio) between two images [Friston et al., 1995a, 1996; Ashburner et al., 1997]. By assuming smooth images and performing a Taylor's expansion of the object image, a least-squares solution can be found for the transformation matrix. This method is only valid when the movements are small relative to the smoothness of the images. SPM also permits reslicing, using linear or sinc interpolation. FIASCO uses a nonlinear optimization technique to estimate the amount of movement necessary to align each image [Eddy et al., 1996b]. Image realignment is done in the Fourier domain. The method used in AFNI is detailed in the user manual, and a reference for a similar method by Irani and Peleg [1991] is provided. AFNI uses an iterative routine to minimize the voxel-to-voxel differences to align two images. Nearest-neighbor, linear, and cubic interpolation are available for resampling. MEDIMAX recently added an iterative optimization technique to perform image realignment. Stimulate does not have an image realignment routine.

The most widely accepted approach for spatial normalization is to transform image data to the stereotaxic Talairach and Tournoux [1988] coordinate system. This was originally proposed by Fox et al. [1988]. Implementation of this coordinate system varies between software packages. SPM uses a linear affine and a nonlinear spatial deformation to transform an image to a template image [Friston et al., 1996]. AFNI's method is based on a piecewise affine transformation to stereotaxic coordinates [Cox, 1996]. Stimulate, MEDx, and the Yale package do not currently provide a published or detailed explanation of the methods utilized to transform images to Talairach-Tournoux space. Both AFNI and Stimulate have extended the number of landmarks to include the cerebellum in the coordinate system, which is not available in Talairach and Tournoux [1988]. Spatial normalization is not available in FIT, MEDIMAX, or FIASCO.

For most of the packages, spatial smoothing is performed by convolving the image volume with a Gaussian filter. Several of the packages provide additional filtering options. Recent research has explored alternative ways to apply smoothing kernels to image data [Poline and Mazoyer, 1994a,b; Siegmund and Worlsey, 1995].

Statistical models

SPM employs the general linear model (GLM) for detection of activations [Friston et al., 1995b]. The analysis of fMRI time-series data is an extension of the model SPM uses to analyze PET data. The GLM assumption of independence of the data is violated in fMRI time-series data due to the temporal correlations observed. To adjust for this violation, SPM recalculates the variance estimators on the data after they have been smoothed. The degrees of freedom are then adjusted to include the effects of smoothing. An assumption is made that the distribution of temporal correlation is known or can be estimated. This extension has been worked out by Worsley and Friston [1995]. AFNI employs the correlation method [Bandettini et al., 1993; Cox et al., 1995] as an option in assessing functional activation. The issue of serially correlated data is addressed by reducing the degrees of freedom used to calculate the significance threshold [Cox et al., 1995]. Stimulate, FIT, Yale, and MEDIMAX have not addressed the problem of temporal correlations in the data.

Correction for multiple comparisons

If a goal of fMRI analysis is to draw conclusions about which brain regions are activated during task performance, then it is essential to correct for the large number of statistical comparisons performed. Correcting for multiple comparisons is complicated by the nonindependent nature of the image data. Therefore, Bonferroni corrections that consider voxels as if they were independent observations are too conservative. Friston et al. [1991] used the Gaussian random field theory to correct for multiple comparisons while controlling for the nonindependence of the data for 2D processes. Worsley et al. [1992] extended the test for three or more dimensions, using a more straightforward procedure. In Friston et al. [1995c], the equation used in SPM96 for assigning probability values in statistical inference is given [see also Poline et al., 1995]. AFNI relies on Monte Carlo simulation or a Bonferroni correction to address the multiple comparison problem. Stimulate utilizes a Bonferroni correction via an auxiliary package [Xiong et al., 1995]. MEDx offers a choice of a Bonferroni correction or the method detailed in Worsley et al. [1992]. FIASCO determines the significance threshold based on cluster size [Forman et al., 1995]. Statistical inference is premature in the current versions of FIT, MEDIMAX, and Yale.

DISCUSSION

After examination of several of the available fMRI analysis software packages, we concluded that although the packages were neither comprehensive nor interchangeable, each contained many useful features. The choice of which package to adopt would depend on the interests and goals of each laboratory. Our main objective for this evaluation was to delineate and summarize individual features of fMRI software packages to aid laboratories in the early stages of developing an fMRI research effort.

The decision of which package to choose will depend on the programming resources of the users. The amount of time the laboratory members are willing to commit to understanding and choosing between particular routines (e.g., linear vs. sinc interpolation in resampling after registration) is an important consideration. Another consideration is the issue of file input/output. All of the packages included in this report, except MEDx, require processing to convert fMRI data from native format to a form recognizable by each software package. MEDx has an automated routine, bypassing the file conversion process. Packages that require only minimal changes such as small descriptor files to read native file formats are preferable to creating multiple versions of the same data set.

The GUI-based packages have particular advantages for users without programming skills, as well as large fMRI laboratories where many researchers will use the software package. GUI allows the user to choose from multiple options, and to point and click to invoke the routines. Prerequisite computer skills are minimal, and the best way to learn how to use this type of package is usually to read the manual and have it in hand for guidance while learning the program's options. One disadvantage of GUI-based packages is that the user typically has to type in several redundant pieces of information for each analysis (e.g., each subject file, the duration of each stimulation, and control condition). It is also difficult to add routines to extend the program's functionality. Another disadvantage is that it is often difficult to see how particular operations are calculated. Users with extensive programming skills may appreciate the flexibility of a modular organization, allowing a package to be tailored to individual needs by interchanging routines from other packages or creating and adding routines.

AFNI may be a good choice for laboratories with multiple users and particularly those without programming interests. We found AFNI to be a user-friendly package. The layout of the program is appealing. The instructions in the user manual are straightforward for formatting our raw image data into an AFNI .BRIK data set. Many of the formulas used in the calculations are available in journal articles or as appendices in the manual. Further explanations for some of the functions can be found in Cox and Hyde [in press]. A unique feature available with AFNI is a manual for programming-literate users, detailing how to create external software packages, referred to as "plugins," that may be read and run by AFNI. AFNI also has several auxiliary programs such as several ANOVA routines, routines to transfer data between different computer systems, programs to convert data from 2D to 3D, and vice versa. A disadvantage of AFNI is that it does not utilize a nonlinear deformation in its spatial normalization algorithm. Stimulate has a layout similar to that of AFNI, but does not contain many of the basic features.

SPM has been widely used for PET analysis and has also become a popular choice for fMRI analysis. Several published journal articles describe many of the functions of SPM. The SPM website is a very good place to start, as papers may be downloaded from the site in addition to many other sources of information about SPM. The SPM E-mail discussion group is also a good place to learn about the package. Routines are available for spatial transformation, realignment, multimodality image registration, linear and nonlinear spatial normalization, spatial and temporal smoothing, and parametric analysis techniques.

A program with a hierarchical organization, such as FIASCO, may provide more options for laboratories in which the users would like more control and understanding of individual routines. FIASCO provides the flexibility of interchanging modules. Therefore, data-processing steps may be compared with other packages at each step or replaced with procedures from other packages. Detailed output of plots and statistics is intended to provide the user with an evaluation of the data quality for each step of data processing. This will allow users to easily compare the output of individual routines with that of other programs. FIASCO also contains routines to correct for baseline miscalibration, scan line mistiming, signal drift, and detrend variability, although these methods have not been published. However, FIASCO does not contain image manipulation tools.

MEDx has an appealing advantage for many researchers in that it provides full technical support. Another advantage of MEDx is that it can directly read 20 different file formats without the conversion steps necessary in other packages, which involve storing multiple versions of data sets. It also has a very extensive user's manual. The package contains some widely used routines such as the AIR algorithm [Woods et al., 1992] and SPM [Friston et al., 1991] within the package. The package also contains many data-processing options. Disadvantages are the cost of the package and the unavailability of the source code for the entire package.

Some of the available packages have been through multiple versions and have worked out many of the bugs. New routines are often added with each upgrade. Many laboratories create their own packages, although this is not a feasible option for all research groups, since development requires a great deal of programming time. It also may take longer to fully test the program when only one group is using the software. Papers published in peer-reviewed journals are an important means for potential software users to assess the validity of routines. This will also facilitate comparison across routines to determine which method is best.

Several choices of software packages are available for the analysis of fMRI data. The list of choices will probably become larger as the field continues to grow and with new laboratories developing research efforts. In deciding which packages are best for a given research group it would be useful to have more than one package up and running, at least in the early stages of a developing laboratory. This would provide opportunities for comparison between packages to get a feel for alternative routines. Furthermore, more than one package may be necessary to complete all processing and analysis steps.

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