

# 3D segmentation of glioma from brain MR images using seeded region growing and fuzzy c-means clustering

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## Declaration

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## Approval Sheet

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## **Abstract**

This thesis presents two algorithms for brain MR image segmentation. The images used are axial MR images of the human brain. The images show a glioma. The objective is to segment the tumour and edema surrounding it from the images. Initially the images are pre-processed by contrast adjustment. Segmentation is performed by two algorithms: seeded region growing and fuzzy c-means clustering. After the images are segmented, the volumes of the segmented regions are measured. The segmentation is done in MATLAB. Finally the results are rendered in 3D in AMIRA.

## **Nomenclature**

MRI: Magnetic Resonance Imaging

FCM: Fuzzy C-Means

CSF: Cerebrospinal fluid

CNS: Central Nervous System

MRF: Markov Random Field

ANN: Artificial Neural Network

GVF: Gradient Vector Flow

FoV: Field of View

ST: Slice Thickness

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# Chapter 1

## Introduction

### 1.1 Background

Brain image segmentation is the process of separating diseased brain tissue from normal tissue. MRI is an important technique to detect such abnormal tissues in the brain. It provides high contrast images of the brain. MRI takes multiple 2D cross-sections to create a 3D structure. In brain MRI analysis, image segmentation is commonly used for measuring and visualizing the brain's anatomical structure. This analysis is used for studying changes in the brain, surgical planning, image-guided interventions, etc. In the case of brain tumours, early detection is crucial. Segmentation of subcortical features from brain images is important for detecting abnormal brain patterns [1].

### 1.2 Motivation

Medical image segmentation is usually done manually by trained experts. These specialists observe the images and draw the boundaries manually on the images. With advancements in scan techniques, large amounts of images need to be segmented. Manual segmentation is too cumbersome and time consuming. It has also been observed that manual segmentation is subject to operator bias [2]. Therefore, there is an increasing need to introduce automation in medical image segmentation. Automated segmentation can be used as a first step before further manual segmentation, if required. By incorporating manual and automated segmentation, we can develop segmentation techniques which are faster and reliable.

### 1.3 Contribution of the thesis

The primary objective of the thesis is the segmentation of brain MR images. The tumour under consideration is a glioma. The tumour and edema surrounding it have been segmented out. Two segmentation algorithms have been used: seeded region growing and fuzzy c-means (FCM) clustering. In region growing, segmentation is done in 3D directly; in FCM it is done separately for each 2D image, and all the results are combined. Volume measurements are made for the segmented regions and both the techniques are compared.

### 1.4 Organization of the thesis

Chapter 2: Overview of brain anatomy

Chapter 3: Describes main types of brain tumours

Chapter 4: Discusses brain MR image segmentation algorithms

Chapter 5: Algorithms and results

Chapter 6: Summary, conclusion, and future work

## **Chapter 2**

### **Brain anatomy**

#### **2.1 Introduction**

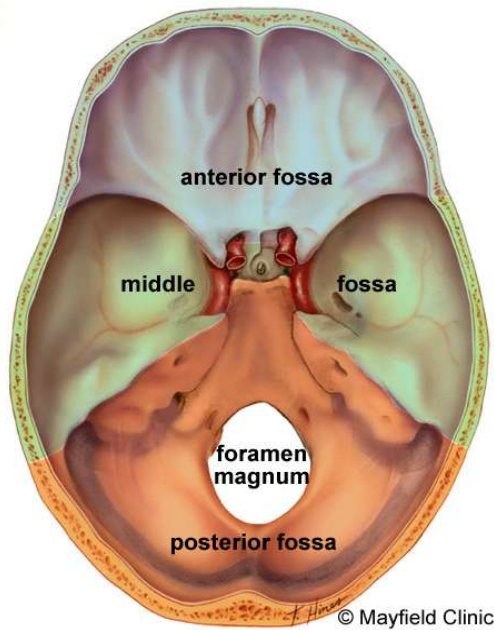
The brain receives information from the outside world through the senses. This information is processed and interpreted in the brain. The brain controls various body functions. The brain also stores the information thus giving rise to memory. Many functions of the body such as thought, movement, speech and various other responses are all controlled by the brain. Heart rate, breathing and the functioning of many organs are also controlled by the brain.

##### **2.1.1 Nervous system**

The nervous system has two parts: central and peripheral nervous systems. The central nervous system (CNS) consists of brain and spinal cord. The peripheral nervous system (PNS) consists of spinal nerves and cranial nerves. Spinal nerves originate from the spinal cord and cranial nerves originate from the brain. Autonomic nervous system, which is part of PNS, regulates various functions like breathing, heart rate, digestion, and secretion of hormones.

##### **2.1.2 Skull**

The skull is a structure of bone which encases the brain. Its purpose is to act as a protective covering for the brain. The skull is composed of eight bones. These bones fuse together along suture lines. The bones that make up the skull are the temporal, parietal, frontal, ethmoid, occipital, and sphenoid. The face is composed of 14 paired bones. These are the maxilla, zygoma, nasal, palatine, lacrimal, inferior nasal conchae, mandible, and vomer. The skull has three distinct areas: anterior fossa, middle fossa, and posterior fossa. At the base of the skull, there are holes called foramina. Arteries, veins, and nerves exit the brain through the foramina.



**Figure 1 Anterior, middle, and posterior fossae**

## **2.2 Brain anatomy**

The brain consists of three parts: cerebrum, cerebellum, and brainstem.

**Cerebrum:** It is the largest part of the brain. It consists of two hemispheres. It performs functions that require higher intelligence. It performs the interpretation of sensations such as vision, hearing, etc. It also is involved in functions such as speech, logic, learning, movement, emotions, etc.

**Cerebellum:** It is located beneath the cerebrum. Its main function is in the control of muscles, and maintaining balance and posture.

**Brainstem:** It consists of midbrain, pons and medulla. It connects the cerebrum and cerebellum to the spinal cord. It controls automatic functions of the body. These functions include breathing, heart rate, maintaining body temperature, wake and sleep cycles, digestion, coughing, and swallowing. Of the twelve cranial nerves, ten originate in the brainstem.

The surface of the cerebrum contains many folds. This folded structure is called the cortex. The cortex contains about 70% of all nerve cells. The cortex is coloured grey-brown because of the nerve cell bodies. This region is called grey matter. Beneath the grey matter is the white matter. White matter consists of axons. Axons are the long part of the nerve cells. The numerous folds in the cortex increase the surface area. This allows more neurons to fit into the cortex, thus improving the ability to perform higher functions. Each fold in the cortex is called a gyrus, and each groove between folds is called a sulcus.

The right and left hemispheres of the brain are connected by the corpus callosum. It is composed of a bundle of fibres. Its function is to deliver messages from one side to the other. Each hemisphere of the brain controls the other side of the body. Speech, comprehension, thinking, etc. are controlled by the left hemisphere, while creativity, spatial ability, artistic skills, etc. are controlled by the right hemisphere. The left hemisphere is dominant in the control of language and hand use in about 92% of people.

### **2.2.1 Brain lobes**

The brain is divided into different lobes by numerous fissures. Four lobes are present in each hemisphere: frontal, temporal, parietal, and occipital. The lobes of the right and left hemispheres are related by many complex mechanisms. The functions of the lobes are as follows [3]:

#### Frontal lobe

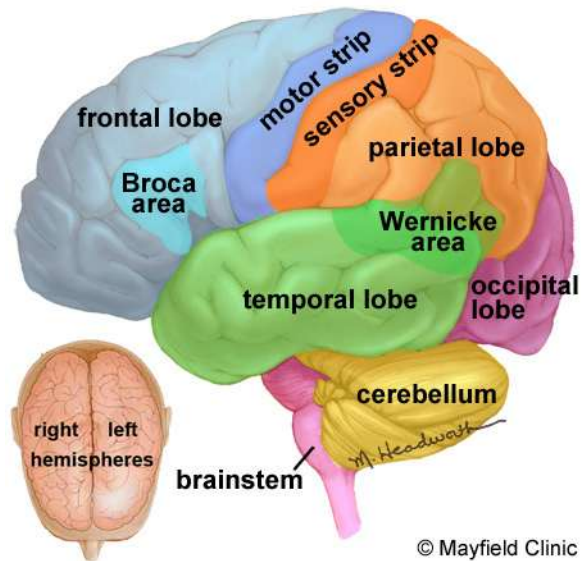
- Emotions, behaviour
- Problem solving, reasoning, planning
- Writing, speaking
- Movements
- Self-awareness, intelligence

#### Parietal lobe

- Language interpretation
- Sensing temperature, pain, touch
- Visual and spatial perception

#### Temporal lobe

- Memory
- Hearing
- Organization, sequencing



**Figure 2 Brain regions**

### 2.2.2 Deep structures

**Hypothalamus:** It is situated at the base of the third ventricle. It is involved in the control of the autonomic nervous system. It regulates behaviours such as hunger, thirst, sleep, sexual response, body temperature, blood pressure, emotions, and secretion of hormones. [4]

**Pituitary gland:** It is situated at the base of the skull in the sella turcica which is a small case of bone. The pituitary stalk connects the pituitary gland to the hypothalamus. It is called the master gland as it controls the other endocrine glands in the body. It secretes hormones that regulate sexual development, bone and muscle growth, and fight disease.

**Pineal gland:** It is situated behind the third ventricle. It secretes melatonin which regulates internal clock of the body and the circadian rhythm.

**Thalamus:** It regulates all information that comes and goes to the cortex. It is involved in pain, sensation, memory.

**Basal ganglia:** It consists of caudate, putamen, and globus pallidus. It is involved in the coordination of fine movements.

**Limbic system:** It controls emotions, learning and memory.

### **2.2.3 Cranial nerves**

The brain communicates with the rest of the body through the spinal cord and cranial nerves. There are twelve pairs of cranial nerves. Ten pairs originate in the brainstem. The remaining two pairs, which are for smell and vision, originate in the cerebrum.

### **2.2.4 Meninges**

Meninges consist of three layers: dura mater, arachnoid mater, and pia mater. The dura mater lines the inside of the skull. It is a strong and thick layer which itself is composed of two layers, periosteal and meningeal dura. The dura has many compartment-like folds. Falx and tentorium are two important dural folds. Falx separates the two brain hemispheres. Tentorium separates the cerebrum from the cerebellum. The arachnoid matter is a membrane that covers the entire brain. It is thin and web-like and is made of elastic tissue. Subdural space is the space between the dura and arachnoid membrane. The pia mater consists of many blood vessels that go deep into the brain.

### **2.2.5 Ventricles and cerebrospinal fluid**

Ventricles are hollow fluid-filled cavities in the brain. Choroid plexus which is located inside the ventricles is involved in the production of cerebrospinal fluid (CSF). CSF is a clear, colourless fluid which flows within and around the brain and spinal cord. It acts as a cushion to protect it from injury. It is continuously absorbed and replenished. There are two ventricles located deep in the cerebral hemispheres called the lateral ventricles. CSF is recycled by arachnoid villi which are located in the superior sagittal sinus. A balance is always maintained between the amount of CSF absorbed and the amount produced. If there is a disruption or blockage in the system, it can cause a build-up of CSF, which may result in enlargement of ventricles (hydrocephalus) or collection of fluid (syringomyelia).

### **2.2.6 Blood supply**

Blood supply in the brain is regulated by neurons and astrocytes. Two paired arteries supply blood to the brain, the internal carotid arteries and the vertebral arteries. The cerebrum is supplied by the internal carotid arteries. The cerebellum, the brainstem, and the underside of the cerebrum are supplied by the vertebral arteries. The basilar artery and the internal carotid artery come together at a structure at the base of the brain called the Circle of Willis [5]. A sort of communication happens between the arteries at this place. If one of the major vessels gets blocked, it is possible for collateral blood flow to come across the Circle of Willis and

prevent brain damage. Thus this meeting of the two arteries is a vital safety feature of the brain.

Venous circulation in the brain follows a different mechanism as compared to the rest of the body. In other parts of the body, for the blood to drain a particular area, arteries and veins must flow together. In the brain there are no pairs of internal carotid veins or vertebral veins corresponding to internal carotid arteries and vertebral arteries. The vein collectors are part of the dura as venous sinuses. These venous sinuses collect blood from the brain and supply it to the internal jugular veins. The internal jugular veins are the only drainage of the brain.

## 2.3 Brain cells

Brain cells are of two types: nerve cells (neurons) and glia cells.

### 2.3.1 Nerve cells

A nerve cell (neuron) consists of three parts: cell body, dendrites, and axon. Neurons transmit information in the form of electrical and chemical signals. There is a tiny gap in between adjoining neurons called synapse. At the end of neurons, there are sacs containing neurotransmitters. These sacs open into the synapse. The neurotransmitter molecules transmit the signal across the synapse to reach special receptors on the receiving nerve cell. These receptors stimulate the next nerve cell to continue transmitting the signal. Anatomical and physiological evidence has been found for the existence of discrete populations of interconnected neurons in the brain in the CNS [4].

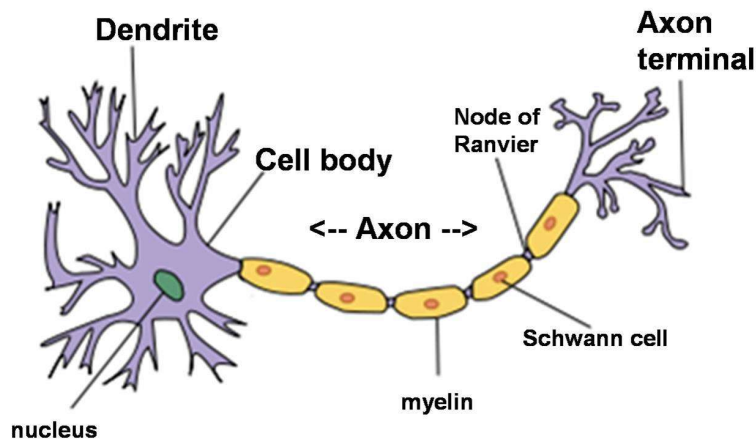


Figure 3 Neuron structure



### **2.3.2 Glia cells**

Glia cells have three functions. They provide nourishment, protection, and structural support to neurons. The number of glia cells is much higher than that of neurons. Most brain tumours originate in glia cells and are consequently called gliomas. Glia cells are of various types. Astroglia or astrocytes supply nutrients to neurons, and regulate the blood-brain barrier. They also digest dead neurons and pathogens. Oligodendroglia cells provide insulation to neurons. Ependymal cells line the ventricles and secrete CSF. Microglia cells digest dead neurons and pathogens.

## Chapter 3

### Brain tumours

#### 3.1 Classification of brain tumours

A tumour is a tissue that grows abnormally due to uncontrolled cell division. Normally cell growth occurs in a controlled manner. A tumour is also called a lesion or neoplasm. Brain tumours are broadly classified into two types:

##### 3.1.1 Primary brain tumours

They originate from brain cells. These tumours may be benign or malignant. Benign tumours grow slowly and have distinct boundaries. They usually do not spread to other brain regions. Benign tumours can be life-threatening if they are situated in sensitive areas of the brain. Malignant tumours grow fast and have irregular boundaries. They spread to other brain regions. Although they are sometimes referred to as brain cancers, malignant tumours are technically not defined as brain cancers because they do not spread to organs outside the brain and spinal cord.

##### 3.1.2 Secondary brain tumours

They spread from other parts of the body to the brain. The cancer cells are carried in the blood stream to the brain. The most common secondary brain tumours start from lung and breast cancers. They are also called metastatic brain tumours.

All brain tumours (benign, malignant, or metastatic) are life-threatening. As the brain is enclosed within the skull, it cannot accommodate a growing tumour tissue. As a result, the tumour compresses and displaces normal brain tissue. Sometimes, it causes a blockage of cerebrospinal fluid (CSF) that flows around and through the brain. This blockage increases intra-cranial pressure causing enlargement of ventricles (hydrocephalus). Some brain tumours result in swellings called edema.

The World Health Organization (WHO) has a classification system for brain tumours [6]. Tumours are classified on the basis of their cell type; if they consist of more than one type of cell, it is a mixed tumour. Tumours are classified by their cell type and grade.

Cell type: This classification is based on the cells from which the tumour starts. Brain tumour can start from nerve cells or from glia cells. Around half of primary brain tumours start from glia cells. These are called gliomas.

Grade: This classification is based on the appearance of tumour cells under a microscope. The grade of a tumour indicates its aggressiveness. A tumour usually has cells of different grades and can also change grades as it grows. Tumour cells which look similar to normal cells are called differentiated cells while those which look different are called anaplastic cells.

### **3.2 Causes**

The exact cause of the origin of tumours is still not known. Some factors have been observed to increase the chances of a tumour. Some of these common risk factors are prolonged exposure to industrial solvents, pesticides, and other chemicals. Secondary brain tumours are caused by tumours from other parts of the body spreading to the brain.

### **3.3 Symptoms**

Tumours affect the brain in many ways. They can increase intracranial pressure leading to compression of normal tissue. Symptoms of the tumour depend on its type, size, and location in the brain. Headaches, seizures, dizziness, vision problems, weakness on one side of the body etc. are some common symptoms of brain tumours. An increase in intracranial pressure can cause headaches, nausea, vomiting, etc. Frontal lobe tumours usually cause behavioural and emotional changes, memory loss, paralysis on one side of the body, and loss of vision. Parietal lobe tumours may cause impaired speech, lack of recognition, and spatial disorders. Occipital lobe tumours can cause loss of vision. Temporal lobe tumours can cause impaired speech and memory loss. Brainstem tumours may cause difficulty in speaking and swallowing, drowsiness, hearing loss, muscle weakness on one side of the body, vomiting, etc. Pituitary gland tumours can cause increased secretion of hormones, stop in menstruation, abnormal secretion of milk, etc.

### **3.4 Diagnosis**

Initially, a complete physical examination is performed and the medical history of the patient and the patient's family are checked. A neurological exam is performed to check the patient's mental status and memory, nerve function, muscle strength, coordination, reflexes, and response to pain. Additional tests such as audiometry, visual field acuity test, endocrine evaluation, etc. may be done if required. Cerebrospinal fluid can be examined by performing a lumbar puncture.

### **3.4.1 Imaging tests**

Computed Tomography (CT): A series of x-ray images are taken from different angles to generate multiple 2-D views of the brain. A computer processes these individual slices to create a 3-D image. A dye may be injected into your bloodstream as a contrast agent.

Magnetic Resonance Imaging (MRI): MRI uses a magnetic field and radio-frequency waves to form an image of soft tissues. It acquires the image in the form of 2-D slices. A dye may be used as a contrast agent. Contrast agents can selectively bind to abnormal vessels within a tumour [6]. MRI is non-invasive and is often used for evaluating brain tumours.

### **3.4.2 Biopsy**

In a biopsy, a small amount of tumour is removed and examined by a pathologist under a microscope. The sample can be taken by an open surgery or by a needle biopsy. In a needle biopsy, a small hole is drilled in the skull, and a hollow needle is inserted through it into the tumour to remove a tumour sample. If the tumour is located in a critical region, a stereotactic biopsy is performed. Volume and number of stereotactic biopsy samples are limited and require specialized histopathological approach [7].

## **3.5 Treatment**

Treatment options vary depending on the type, grade, size, and location of the tumour.

### **3.5.1 Observation**

Benign tumours may be observed using MRI scans. As benign tumours grow slowly, observation is continued until it reaches a stage where surgery is necessary. Old patients who have other health conditions are more commonly subject to observation.

### **3.5.2 Medication**

Medication is used to treat effects of brain tumours. Corticosteroids are used to reduce swelling or inflammation around the tumour. Furosemide or mannitol is used to treat edema and intracranial pressure. Anticonvulsant medications are used to prevent or control seizures.

### **3.5.3 Surgery**

Surgery is preferred if it is possible to reach the tumour without damaging any vital brain regions. A craniotomy is performed to open the skull and remove the tumour. If the tumour cannot be completely removed without damaging vital brain regions, only a part of the tumour is removed. The remaining tumour cells may be treated with other methods like radiation or chemotherapy. A partial removal of the tumour may also relieve many of the symptoms.

### **3.5.4 Radiation**

High-energy radiation is directed towards the tumour cells to destroy them. It destroys the cell DNA rendering the cells unable to reproduce. The radiation is given such that tumour cells get maximum dose and normal cells get minimum dose. Radiation can be given in two ways: external and internal.

External beam radiation is delivered from outside the body. Stereotactic radiosurgery (SRS) delivers a high dose of radiation in a single session. Fractionated stereotactic radiotherapy (FSR) delivers lower doses of radiation over many sessions. Whole brain radiotherapy (WBRT) delivers the radiation dose to the entire brain.

Internal radiation is delivered from inside the body. A craniotomy is performed to remove the tumour. After it is removed, a radioactive material is placed in the tumour cavity. The radiation from this material will destroy tumour cells that may remain in the surrounding tissue.

### **3.5.5 Chemotherapy**

Chemotherapy drugs can be administered orally, intravenously, or in the form of a wafer placed in the tumour. These drugs disrupt cell division of tumour cells. Healthy cells are also affected, but they can repair themselves. It is given in doses with rest periods allowed in between. It is used mainly for high-grade tumours.

## **3.6 Glioma**

Gliomas are brain tumours that originate in the glial cells. There are different types of gliomas based on the type of glial cells they originate from. There are different types of glial cells. The functions of various types of glial cells are given below:

Astrocyte: Transport nutrients to neurons and hold neurons in their positions

Oligodendrocyte: Provide insulation to neurons

Microglia: Digest dead neurons and pathogens

Ependymal cells: Line the ventricles and secrete cerebrospinal fluid

Symptoms of gliomas depend on their location in the brain. Usual symptoms include headaches, weakness, changes in behaviour, seizures, etc. Treatment options are based on the severity of the glioma. Gliomas are classified into different grades as follows:

Grade I – Pilocytic astrocytoma:

- Benign and slow-growing
- Occur usually in children, less commonly in adults

- Appear in cerebellum or brainstem, occasionally in cerebral hemispheres

Grade II – Low-grade glioma:

- Typically occur in young adults
- Occur mostly in the cerebral hemispheres
- May grow into more aggressive tumours, may recur
- Includes astrocytoma, oligodendroglioma, mixed oligoastrocytoma

Grade III – Malignant glioma:

- Grow fast and more aggressively
- Invade nearby tissues with tentacle-like projections
- Symptoms include seizures, headaches
- Includes anaplastic astrocytoma, anaplastic oligodendroglioma, anaplastic mixed oligoastrocytoma
- Recurrence is common

Grade IV – Glioblastoma multiforme (GBM)

- Most aggressive
- Most common primary brain tumour
- Invades other parts of brain with tentacle-like projections
- Recurrence is common

# Chapter 4

## Image segmentation

### 4.1 Introduction

Image segmentation is the process of partitioning an image into mutually exclusive regions such that each region is spatially contiguous and pixels within a region are homogeneous with respect to a pre-defined criterion [8]. An image can be segmented based on similarity or dissimilarity of gray levels. Image segmentation based on gray-level similarity includes methods such as intensity-based threshold, region-based methods like region growing and watershed, fuzzy clustering, etc. Segmentation based on gray-level dissimilarity includes methods such as edge detection, line detection, etc.

### 4.2 Manual and Automated segmentation

Depending on the degree of manual intervention, brain tumour segmentation methods are classified into three types [9]:

#### 4.2.1 Manual segmentation

The boundaries of the required regions are drawn manually and labelled [10]. It is performed by clinicians/radiologists or other experts. The person has to segment each 2-D slice separately. It is tedious and time-consuming. The results of manual segmentation are subjective and show significant intra and inter-rater variability [11]. Despite these problems, manual segmentation is still used as a validation for automatic and semi-automatic segmentation results.

#### 4.2.2 Semi-automatic segmentation

In semi-automatic segmentation, some user interaction is present. User interaction is mainly of three types [12]:

Initialization: Input of parameters, selection of certain pixels from the image, etc. Sometimes, pre-processing of the data is performed by the user.

Feedback: If undesired results are obtained while the process is being executed, it is stopped and resumed after necessary changes are made.

Evaluation: Results are evaluated by the user to check if they are satisfactory or not.

Semi-automatic segmentation is the most widely used of the three degrees of manual intervention.

### **4.2.3 Automatic segmentation**

In automatic segmentation, there is no human interaction. It is very difficult for a computer to match the ability of human vision. However, brain tumour imaging has some advantages. The appearance of the brain is similar in most of the images. There is no temporal component as the brain remains stationary. Also, only one slice is considered at a time. Therefore, the human brain's advantage of analysing 3-D and moving objects does not matter much.

Humans use very high-level visual processing and incorporate specialized information about the domain. This level of intelligence is difficult to replicate in a computer [13].

Currently, fully automatic methods are used only for segmenting extremely large amounts of data, and that too only in research environment.

## **4.3 Unsupervised and Supervised segmentation**

### **4.3.1 Unsupervised segmentation**

It does not use labelled training data. The number of classes is specified by automatically by the training algorithm. The algorithm also assigns physical labels to the clusters [14]. The clustering is done using either an anatomic objective measure or an image-based objective measure. The absence of prior information on shape or intensity makes unsupervised segmentation of brain tumours more difficult [15].

Anatomic objective measure aims to segment the image into anatomically different regions. In brain tumour imaging, it tries to segment the image into tumour, edema and other regions. This algorithm may face difficulties as the anatomic feature may itself consist of two or more distinct parts. A brain tumour may be comprised of different regions. To reduce such problems, pre-processing is done on the image. Two common pre-processing techniques are image inhomogeneity correction and skull stripping.

### **4.3.2 Supervised segmentation**

It uses labelled training data. In the training phase, labelled data is used to learn a model for the clustering. This is followed by the testing phase in which unlabelled pixels are classified based on their intensities. The results of supervised segmentation depend on the training data used. Therefore, accurate training data is required. The number of classes in the result is decided by the operator. The training can be patient-specific or inter-patient. In patient-specific training, the training data is taken from the image to be segmented. In inter-patient training, the training data is taken from different patients. As supervised segmentation uses



training data, it is more effective than unsupervised segmentation. The choice of training data is very important as different training data lead to different results.

#### **4.4 Threshold-based segmentation**

Intensity-based threshold is the simplest method for segmentation. Each pixel is compared with a selected threshold value. Pixels with intensities above and below the threshold are assigned to different classes. If one threshold is used, it gives a binary image. Threshold can be applied in two ways: global and local.

##### **4.4.1 Global threshold**

The threshold is applied for the entire image. It does not consider any local correlation between adjacent pixels. In the presence of noise, it gives very poor segmentation. Changes in intensity inhomogeneity affect the segmentation result adversely.

##### **4.4.2 Local threshold**

Threshold is chosen adaptively in the local region around each pixel. For each pixel, the threshold is calculated using local statistical properties such as the mean intensity value [16] of the neighbouring pixels.

Intensity-based threshold segmentation is usually used as a first step in the segmentation process.

#### **4.5 Region-based segmentation**

Region-based segmentation divides the image into different regions by merging neighbouring pixels with homogeneity properties based on similarity criterion. Region-based segmentation methods include region growing and watershed segmentation. These are also the most commonly used methods for brain tumour segmentation. Some region-based methods merge region growing and region merging techniques.

##### **4.5.1 Region growing**

Region growing extracts a connected region of similar pixels from an image [17]. A seed point is selected within the required region. A homogeneity criterion is determined using similarity in intensity. The segmentation starts from this point. The neighbouring pixels are tested for the homogeneity criterion. If they satisfy it, they are added to the region. Then the process is repeated on the neighbouring pixels of the new region. This method is explained in greater detail in a later section.

#### **4.5.2 Watershed segmentation**

In watershed segmentation, the image is considered as a topological surface. If this surface is flooded with water, the surface would be partitioned into two: catchment basins and watershed lines. The different catchment basins are the different regions into which the image has been segmented, while the watershed lines serve as the boundaries between the regions. Watershed segmentation usually results in over-segmentation. Watershed applications have been widely used in brain MRI segmentation [18].

#### **4.6 Pixel classification**

Pixel classification is done using unsupervised or supervised classifiers. For classification, distance measure can be used as a similarity criterion. The distance measure can be the distance between two vectors or their normalized inner product. Each cluster is represented by its centroid/mean and variance. The classification is optimized using a cost function which maximizes similarity within clusters and dissimilarity between clusters. Unsupervised methods include Fuzzy C-Means (FCM) clustering and Markov Random Fields (MRF). Supervised methods include Bayes and Artificial Neural Network (ANN).

##### **4.6.1 Fuzzy C-Means (FCM) clustering**

This is used when the features that determine homogeneity do not have sharp transitions at region boundaries. Initially, a set of tissue classes is determined. Each pixel is assigned a membership value to each class [19]. The membership function is constrained to have values between 0 and 1. This value indicates the degree to which a pixel belongs to a particular class. This is in contrast to hard clustering where each pixel is assigned to only one class. FCM is explained in greater detail in a later section. Spatial information can be incorporated into the fuzzy membership function to reduce the effect of noise [20].

##### **4.6.2 Markov Random Fields (MRF)**

FCM does not consider spatial information. In brain tumour imaging, spatial information is important because if a pixel is strongly labelled as tumour, its neighbouring pixels will also likely be labelled as tumour. This reduces the effect of noise and overlap of clusters [21]. To represent these complex dependencies, MRF and Conditional Random Fields have been used. CRF variants like Discriminative Random Fields (DRF) and Support Vector Machines (SVM) are coupled with knowledge-based features. MRF models require algorithms that are computationally intensive.

### **4.6.3 Artificial Neural Networks (ANN)**

The features are processed through a series of nodes. Input nodes perform mathematical operations while classification is done at the output nodes. As ANN employs supervised learning, it uses a training data set. In the training phase, parameters in the mathematical operations are optimized so as to minimize prediction error. ANN does not use any parametric distribution, so it is said to be non-parametric. It can model non-trivial distributions. This is useful for brain tumour imaging as brain tumour data may not follow a simple Gaussian distribution. ANN models are complex and time-consuming. Large data sets are required for the training phase. ANN classifiers can use patient-specific training [22]. Some ANN techniques have been developed that do not require patient-specific training [23]. Self-organizing maps (SOM) are a special case of ANN used mainly for visual pattern recognition [24]. SOM is based on topological data of the brain cortex. In the brain cortex, topologically closer neurons respond to the same stimulus. SOM is based on competitive learning.

## **4.7 Model-based segmentation**

Model-based segmentation techniques use a deformable model which changes its contours according to certain properties. The models may have prior statistical information drawn from training datasets. Using a priori information, a continuous model of the required anatomic feature is created. In some cases, statistical information from training datasets is also used. Then the deformation of the model will be constrained by the statistical information.

The deformable model is a propagating interface which deforms under the control of local and global constraints. This propagating interface will be a closed curve in 2D and a closed surface in 3D. Deformable models are especially used in the segmentation of volumetric (3D) data. Deformable models are mainly of two types: parametric and geometric.

### **4.7.1 Parametric deformable model**

Parametric deformable models are also called active contour models or snake models. It can detect lines, edges, and subjective contours. An initial set of contour points is selected somewhere close to the required feature. This contour changes its shape by a process of energy minimization. This is done by iteratively minimizing an energy functional [25]. The functional consists of two components: internal and external energy.

Internal energy is associated with the curve that is being deformed. It imposes a smoothness constraint on the curve. It is a generalization of a Tikhonov stabilizer. External energy is

dependent on the image. External energy minimization moves the curve towards image features. External energy functional has three components: line, edge and termination functional. External energy is positive in homogeneous regions and zero at boundaries. The energy minimization is done iteratively. At each step of the iteration, the points of the contour are updated to their new values.

Conventional segmentation techniques work in a bottom-up manner. Therefore, an error made at an initial step gets carried over to the succeeding steps. Parametric models combine the bottom-up approach with a top-down approach. If any part of the contour comes in contact with the required feature, that part pulls the rest of the curve towards the required feature. This is called scale space property. Parametric models have interactive mechanisms that allow scientists and experts to bring their knowledge to interactively guide the segmentation process [26].

Parametric models do not converge properly if the required feature has concavities in its boundary. Also it is important for the initial contour to be placed near the required feature. To overcome these problems, gradient vector flow (GVF) models [27] and balloon models have been developed. GVF uses spatial diffusion of the gradient of the edge map instead of image gradient as external force.

#### **4.7.2 Geometric deformable model**

Geometric deformable models are based on the curve evolution theory and the level-set model. The model deforms using only geometric measures; it is independent of parameterization. The deformation is coupled with image data. Topological changes are naturally possible. Level set algorithms are computationally expensive.

# Chapter 5

## Experiments & Results

### 5.1 Data

The data set consists of 192 axial brain MR images. They are grayscale images of resolution 512 x 512. They show a glioma. Each slice has a thickness of 6 mm. The Fields of View along x axis and y axis are 120 mm and 50 mm respectively.

Image segmentation was performed on MATLAB platform. 3-D rendering was obtained in AMIRA™.

### 5.2 Contrast adjustment

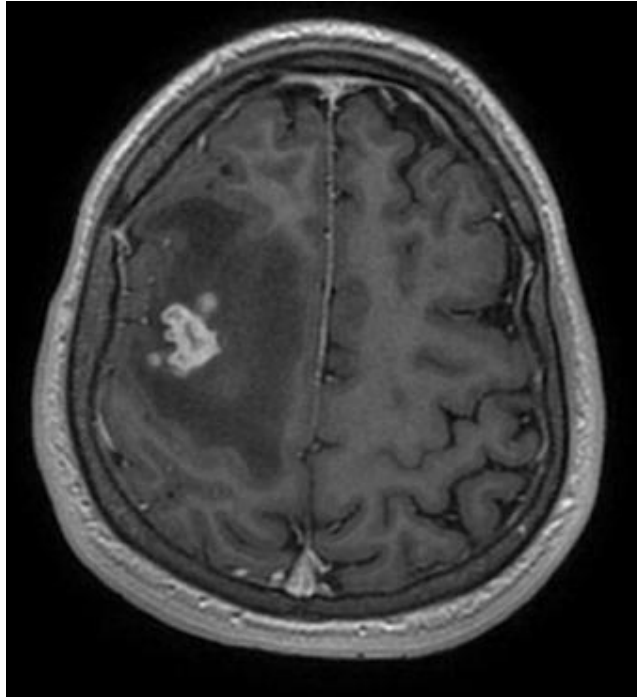
Contrast adjustment improves the contrast of tumour and edema regions with respect to their backgrounds. The pixel intensities of tumour and edema before and after contrast adjustment are given in the following tables.

Tumour	Minimum	Maximum
Original	124	187
After contrast adjustment	170	255

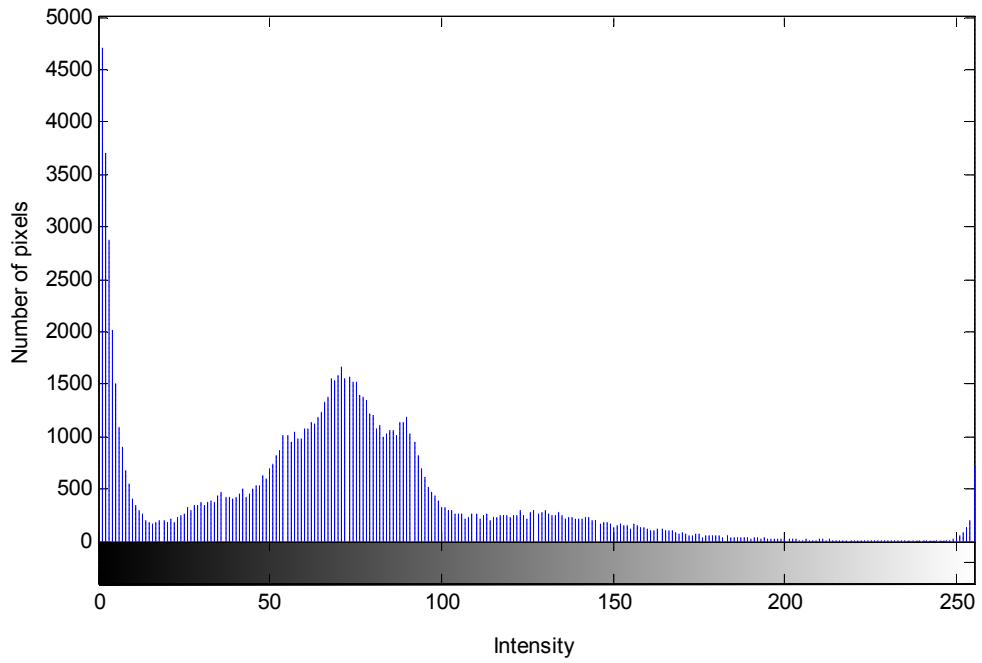
**Table 1**

Edema	Minimum	Maximum
Original	46	64
After contrast adjustment	0	21

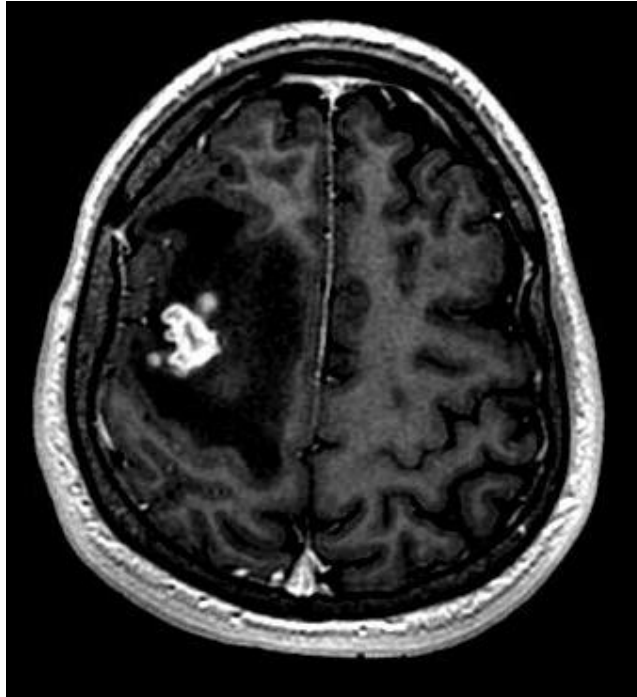
**Table 2**



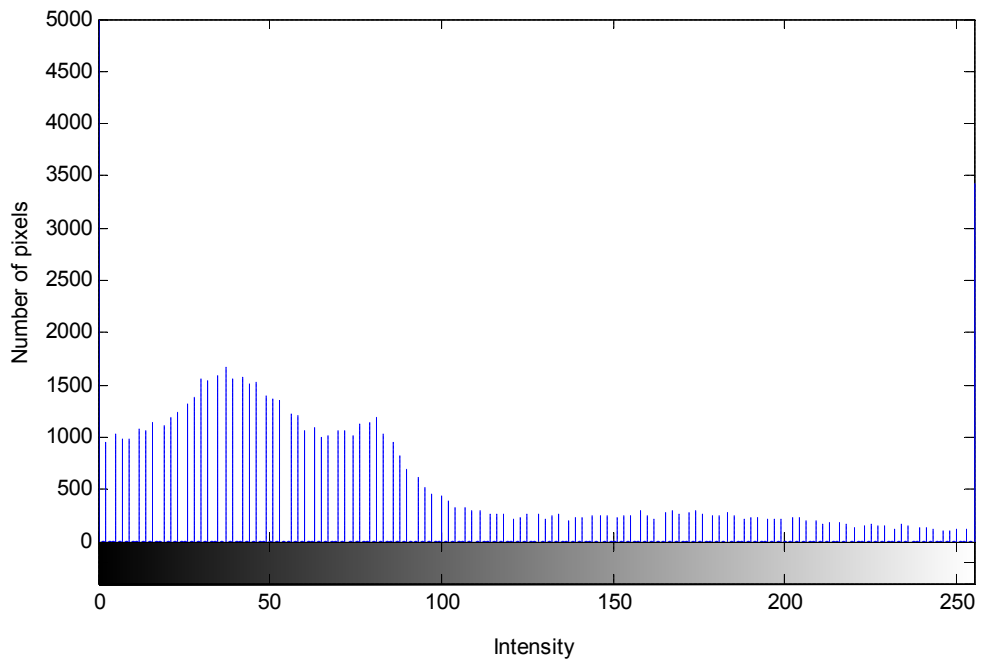
**Figure 4 Sample image from data set**



**Figure 5 Histogram of sample image**



**Figure 6 Image after contrast adjustment**



**Figure 7 Histogram after contrast adjustment**

## **5.3 Region growing**

### **5.3.1 Introduction**

In segmentation techniques such as intensity-based threshold and gradient-based edge detection, the problem was to segment the image by finding the edges around the regions. Region growing method can be considered as the dual problem to this. In region growing, the problem is to find the region themselves. The algorithm starts from a point within the region and grows outwards until it reaches the boundaries of the region. Region growing is effective and less computationally intensive as compared to other non-region based segmentation methods. Seeded region growing has been used in the segmentation of MR images acquired in all three orientations [28].

### **5.3.2 Region characteristics**

The regions that are developed are usually considered to be non-overlapping 2-D areas. Each region is a connected region. It can also be non-simply connected, i.e. it can have holes inside it. However no two regions should overlap or no pixel should belong to more than one region. These definitions of the image regions can vary depending on the application and the kind of image that is being segmented.

### **5.3.3 Seed selection**

The point from which region growing process starts is the seed point. The method used for seed selection depends on the application. If a priori information on the image is available, it may be used to select the seeds. If no a priori information is available, the seed may have to be selected automatically. For this, histogram is used. The pixels which correspond to the strongest peaks on the histogram can be chosen as the seeds [29]. In semi-automatic segmentation, seed selection is performed manually.

To start the process of region growing, any pixel that lies within the required region is selected as the seed. There is a chance that the selected seed point may fall on a pixel that is not characteristic of the region even though it may lie within the region. This happens especially if there is noise. To solve this, multiple seeds are selected. Then the average of their intensities is computed. Any of the points that lie within a certain pre-defined range of the average is chosen as the seed point. This process ensures that the chosen seed point is characteristic of the required region. In case multiple regions need to be grown, different seed



selections have to be performed for each region. This is done if the required region is not a connected region.

#### **5.3.4 Neighbourhood selection**

Selection of neighbouring pixels can be carried out in many ways. One simple way is to use a morphological structuring element and perform a dilation operation on the image. This gives a new structure which consists of the original structure and its immediate neighbouring pixels. From this new structure, the original structure is removed leaving only the neighbouring pixels. Thus the neighbouring pixels alone are selected. If region growing is being carried out in 2-Dimensions, an 8-connected structural element is used for dilation. If it is being performed in 3-D, a 26-connected structural element is used.

#### **5.3.5 Homogeneity criterion**

The set of neighbouring pixels is tested for the homogeneity criterion. Each pixel is tested separately. This criterion is used to determine the similarity of the neighbouring pixels to the region. The homogeneity criterion can be based on the intensity or any other feature of the image [30]. If intensity is used, the average intensity of the current region is calculated. For each neighbouring pixel, the difference in intensity with the average of the current region is calculated. If this difference is less than a certain threshold, that pixel is to be included in the region. If it is higher than the threshold, it is to be excluded. This threshold for the intensity difference depends on the intensities of the pixels in the image.

#### **5.3.6 Termination of region growing**

Initially the pixels surrounding the seed point are tested for the homogeneity criterion. Those that satisfy the criterion are added to the region. Then the same procedure is repeated using the new region. This process is continued iteratively. In any step of the iteration, if none of the neighbouring pixels satisfy the homogeneity criterion, then the region does not grow in that step. It means that none of the neighbouring pixels are similar to the growing region, i.e. the region boundary has been reached. When this happens, the region growing process is terminated. As long as at least one pixel is added to the region, the region growing process must be continued.

### 5.3.7 Results

Tumour and edema are the regions to be segmented from the original MR image. The segmentation of these two regions is done separately. In the original MR images, the tumour lies between the pixel intensities [124, 187] and edema lies between [46, 64]. After contrast adjustment, the tumour lies between [170, 255] and edema lies between [0, 21]. Homogeneity criterion is defined as pixel intensity lying within  $\pm 25$  of the seed pixel intensity. Neighbourhood selection is done using the dilation operation method described earlier. As the segmentation is being done in 3-D, a 3-D structuring element which selects the 26-connected neighbourhood is used.

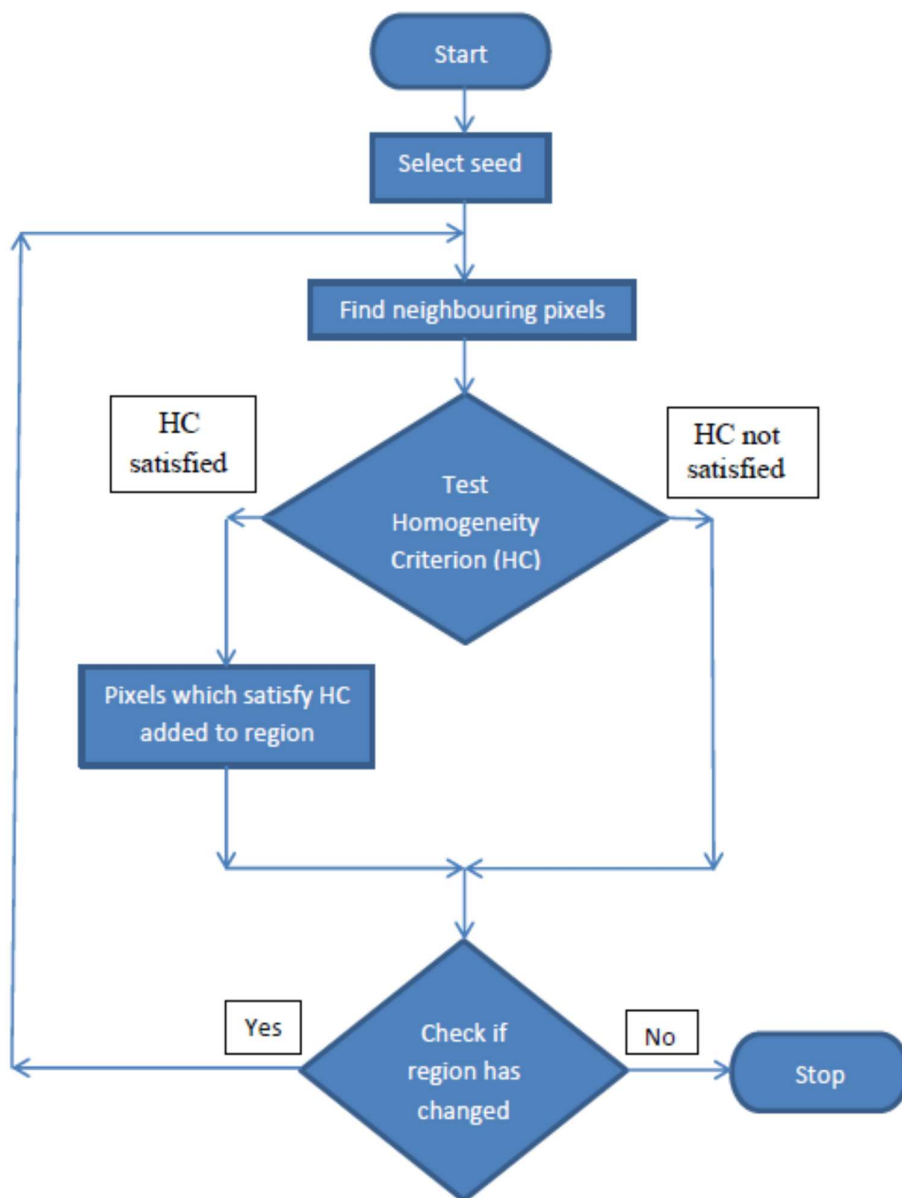
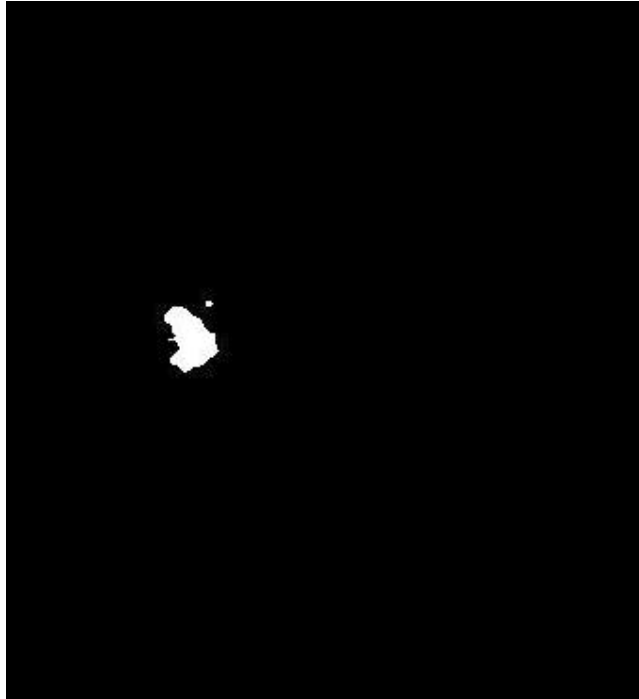
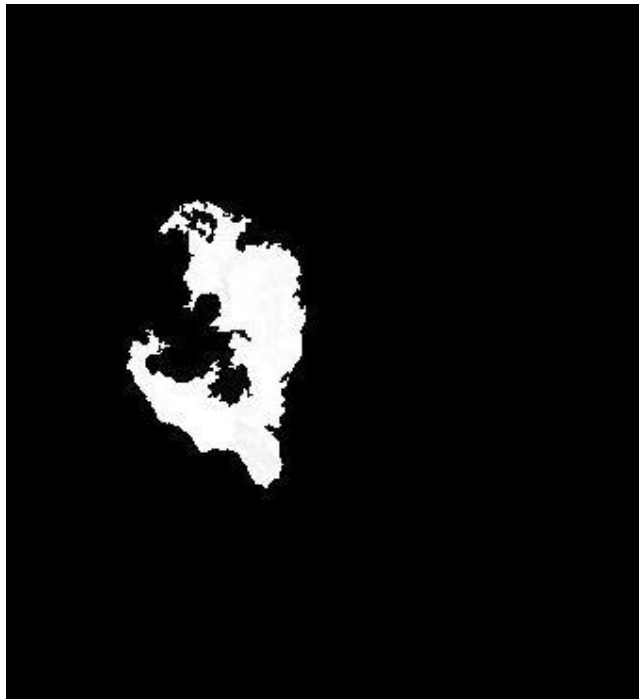


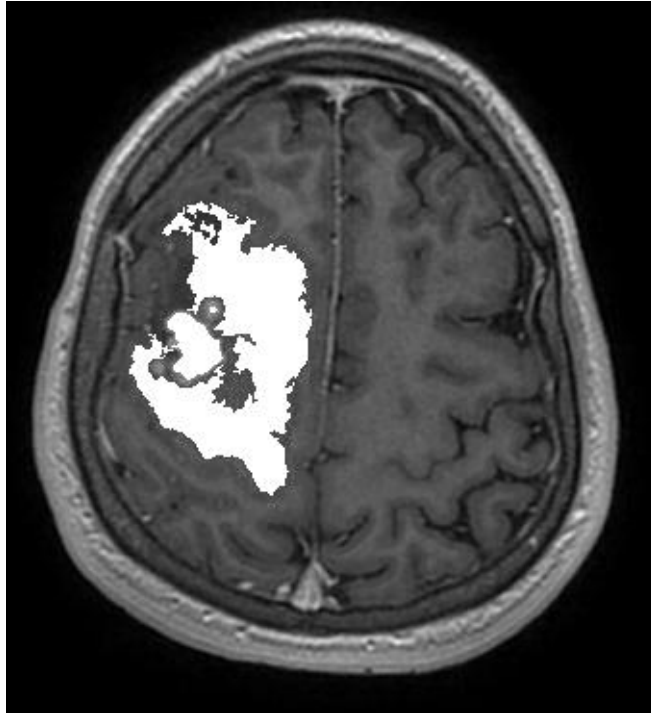
Figure 8 Flowchart of region growing algorithm



**Figure 9 Sample from segmented tumour set**



**Figure 10 Sample from segmented edema set**



**Figure 11 Sample image tumour and edema superimposed on original image**

## **5.4 Fuzzy C-Means (FCM) clustering**

### **5.4.1 Introduction**

Clustering is a method of dividing a data set into different groups or clusters. The data points are assigned to clusters and each cluster can be characterized by a single reference point. This reference point is generally an average of the points in the cluster. In c-means clustering, 'c' reference points are selected. The positions of the reference points and the assignment of the data points to clusters are adjusted and applied over the images to be segmented.

Usually when a data set is divided into clusters, each data point is assigned definitively to any one of the clusters. This is called hard clustering. In fuzzy clustering, each data point is assigned a membership function to each cluster [31]. The membership function of a data point to a cluster indicates the degree to which the data point belongs to that cluster. The membership functions are represented in the form of a matrix called fuzzy partition matrix. Every point has a degree of belonging to each cluster rather than belonging entirely in any one cluster. The fuzzy membership function is constrained to have values between 0 and 1 [32].

### 5.4.2 Algorithm

Fuzzy c-means clustering is based on the optimization of a c-means objective function. The functional which is to be optimized is given as:

$$\sum_{i=1}^n \sum_{j=1}^c w_{ij}^m \|x_i - c_j\|^2$$

Where:  $X = \{x_1, x_2, \dots, x_n\}$  is the set of data points

$C = \{c_1, c_2, \dots, c_c\}$  is the set of cluster centres

$W = w_{ij} \in [0,1], \quad i=1,2, \dots, n, \quad j=1,2, \dots, c$

Fuzzy clustering is done by iterative optimization of the above function with the update of membership  $u_{ij}$  and cluster centres  $c_j$  by:

$$u_{i,j} = \frac{1}{\sum_{k=1}^c \left[ \frac{\|x_i - c_j\|}{\|x_i - c_k\|} \right]^{\frac{2}{m-1}}}$$

The iteration will stop when:

$$\max_{i,j} \{|u_{i,j}^{k+1} - u_{i,j}^k|\} < \delta,$$

where:  $\delta$  is a termination criterion between 0 and 1, and  $k$  are the iteration steps.

This iteration converges to a local minimum or a saddle point  $J_m$ . The algorithm is composed of the following steps:

Initialize  $U = [u_{i,j}]$  matrix.

At step  $k$ , calculate the centre vectors  $c_j$ :

$$c_j = \frac{\sum_{i=1}^N u_{i,j}^m x_i}{\sum_{i=1}^N u_{i,j}^m}$$

Update  $U$ :

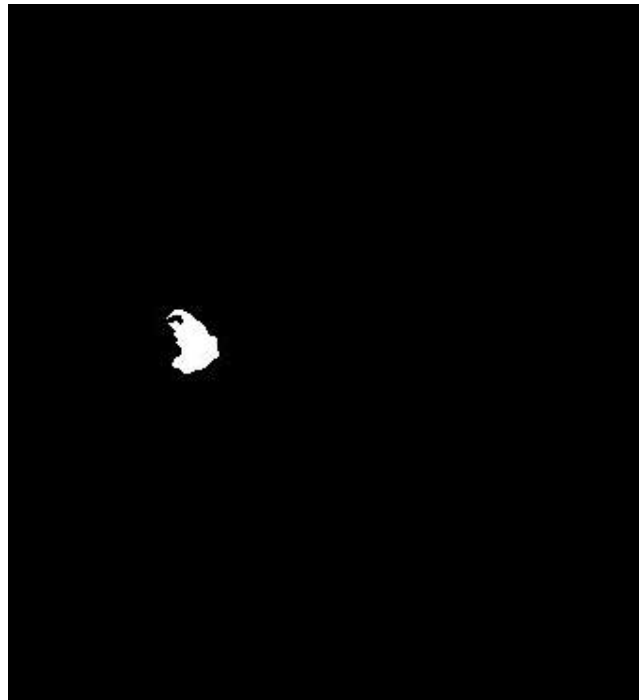
$$u_{i,j} = \frac{1}{\sum_{k=1}^c \left[ \frac{\|x_i - c_j\|}{\|x_i - c_k\|} \right]^{\frac{2}{m-1}}}$$

If  $\|U^{(k+1)} - U^{(k)}\| < \delta$ , then stop the iteration, otherwise, return to step 2.

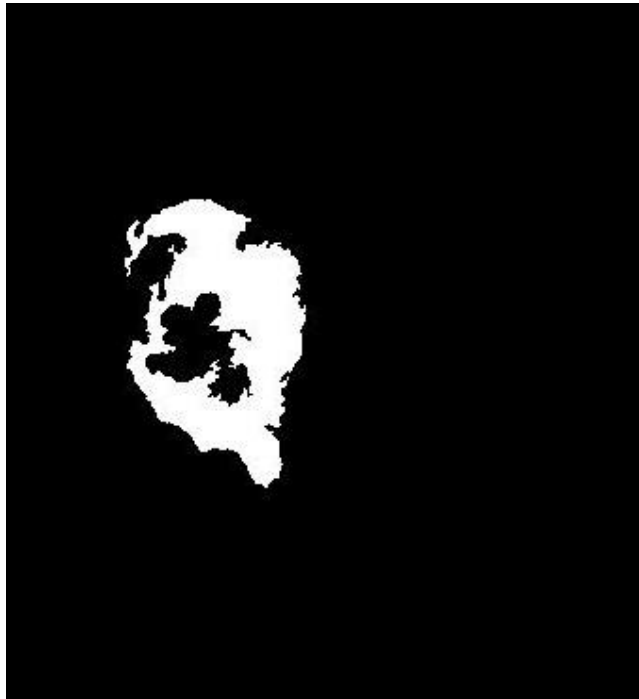
The fuzzy clustering process gives a set of cluster centres and a fuzzy partition matrix. The partition matrix gives the labels of the clusters (usually the centres) and the membership function of each pixel to each cluster.

### 5.4.3 Results

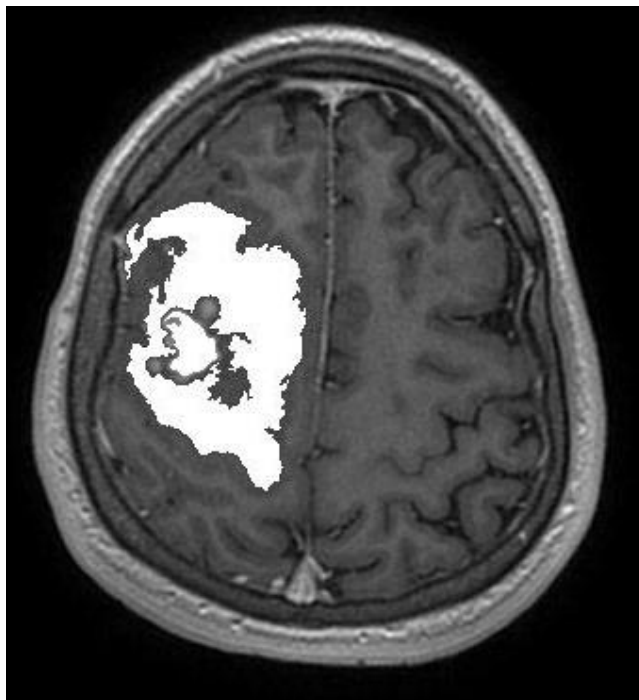
When FCM is applied for brain tumour segmentation, the first step is to define a set of tissue classes. Each pixel is then assigned membership values to the tissue classes. The fuzzy membership functions, constrained to lie between 0 and 1, indicate the similarity of the pixel to each of the tissue classes. If the initialization is done using an approximate estimation of the cluster centres, the algorithm converges faster and the results are more accurate. FCM by itself may not be good enough in brain tumour segmentation as it fails to consider spatial information that is important in brain MR images. FCM, being an iterative algorithm, is also quite time-consuming.



**Figure 12 Sample from segmented tumour set**



**Figure 13** Sample from segmented edema set



**Figure 14** Tumour and edema superimposed on original image

## 5.5 Volume measurements

The volume of a voxel is calculated as:

$$Volume = \frac{FOV_x}{N_x} \times \frac{FOV_y}{N_y} \times S.T$$

where  $FOV_x$  and  $FOV_y$  are the fields of view along x and y axes respectively. S.T is the slice thickness.

$FOV_x$  is 125 mm,  $FOV_y$  is 152 mm and slice thickness is 6 mm. Substituting these values in the equation gives the volume of a voxel as  $0.43488 \text{ mm}^3$ .

From each of the segmented results, the number of pixels which represent the tumour and the edema are counted. The volume of the tumour/edema is found by multiplying the number of pixels with the volume of a voxel. These values are given in the following tables.

Measurements	Tumour	Edema
Number of pixels	9818	189070
Volume ( $\text{mm}^3$ )	4269.65	82222.76

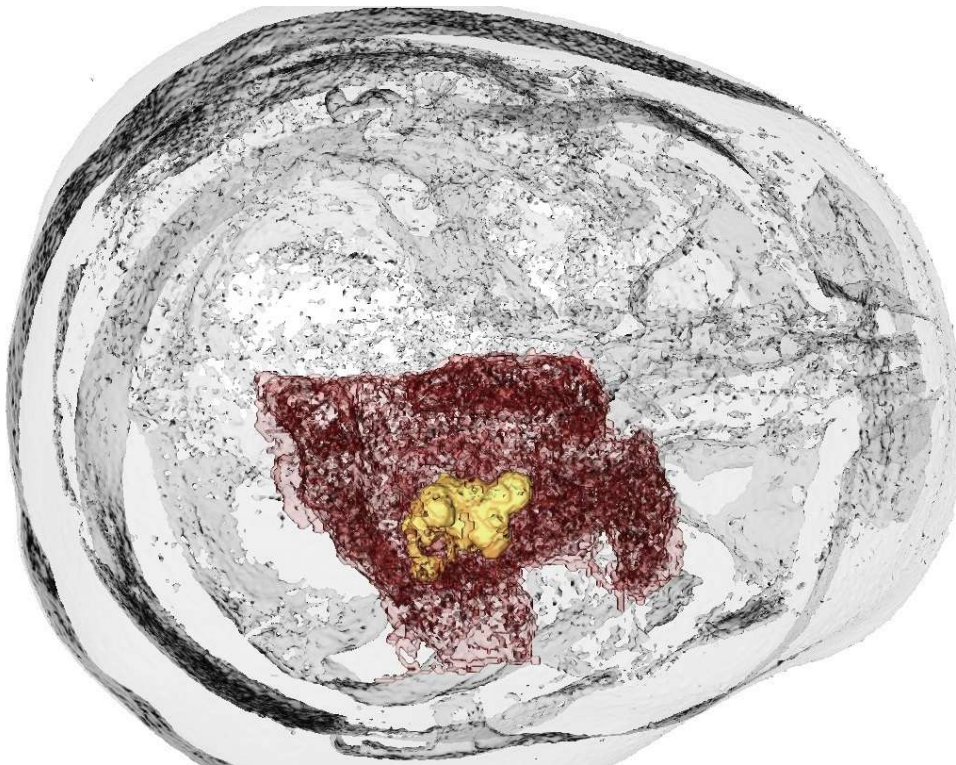
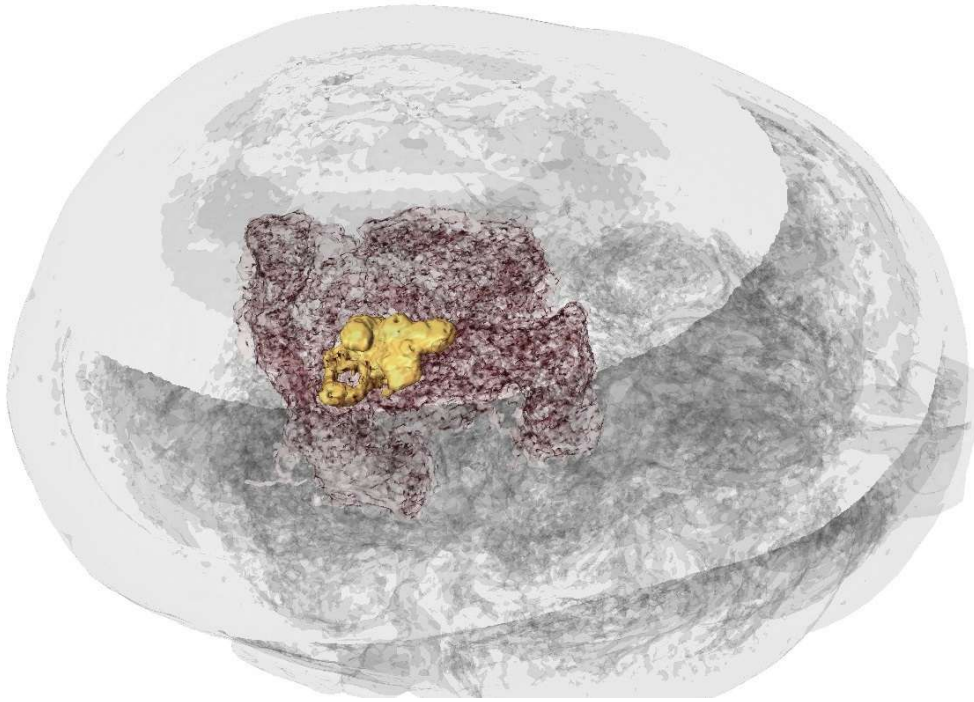
**Table 3 Region growing**

Measurements	Tumour	Edema
Number of pixels	9963	214888
Volume ( $\text{mm}^3$ )	4432.71	93450.49

**Table 4 FCM**

3-D rendering of the brain tumour and edema was obtained using AMIRA software. The tumour, edema, and the original images are loaded as three separate data sets. These 3D views are shown in the following figures.





**Figure 15 3D View**

## Chapter 6

### Summary, Conclusion & Future work

#### 6.1 Summary

The input data set consisted of 192 axial MR images of the human brain. The images showed a glioma. Contrast adjustment was done as a pre-processing step. This improved the contrast of the tumour and edema regions with the background. Then segmentation was performed using two algorithms: region growing and fuzzy c-means clustering. Using both the algorithms, tumour and edema were segmented out from the images. Segmentation was done in MATLAB. Volume measurements were made for the segmented regions. This was done by calculating the volume of a pixel and counting the number of pixels in the required regions. The results were rendered in 3D using AMIRA software.

#### 6.2 Conclusion

Both the techniques used for segmentation are semi-automatic. The segmentation results and the volume measurements made can help in pre-surgical assessment of the tumour. For proper validation of the results, they need to be compared with manual segmentation results for the same data. These algorithms are both quite fast; they take only a few minutes to perform the segmentation, whereas manual segmentation may be rather time-consuming. The results of these techniques are promising, but further research is needed in this field.

#### 6.3 Future work

Adaptive region growing methods have been proposed which use gradients and variances along the curve to develop the region. Modified region growing method incorporates gradient information to remove partial volume effects [33]. In FCM, new algorithms such as enhanced FCM [34] have been proposed to evaluate the neighbourhoods of each pixel as a pre-refinement step.

Although several methods have been developed for brain tumour segmentation, these methods have yet to gain wide clinical acceptance [35]. Consequently further research is needed in this field. One main concern the clinical fraternity has with automated segmentation algorithms is that there is no clear standardization procedure for automated segmentation techniques. Also with these methods, there is a lack of interpretability as compared to segmentation done by trained experts [36].

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