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CleveLabs Laboratory Course System - Student Edition

Polysomnography Laboratory

Introduction

Sleep is a very important part of human life, equally important as water, air, and food. During sleep the body works to strengthen neuronal connections, rebuild cells, and remove byproducts of cell metabolism. A person deprived of sufficient amounts of sleep may develop serious illnesses.

Sleep has mystified scientists trying to understand how a person can be asleep, but at the same time have vivid dreams as if he or she were awake. It was originally believed that during sleep the functions of the body slowed



down. We now know that the opposite is true. The brain, for example, is indeed very active during sleep. At times, when measured by biomedical instrumentation, the brain appears to be awake, even though the person is soundly asleep.

One tool physicians use study sleep is polysomnography (PSG). In this procedure, patients with sleep disorders go into a clinic and sleep there overnight while specialized monitoring equipment measures specific physiological signals. You should be familiar with some of these signals already. The ECG, EEG, EMG, and EOG are all measured during a standard PSG study (Fig 1). In addition to these signals, respiratory monitoring is also critical during a sleep study.

The recorded biopotentials are used to measure and quantify different stages of sleep. As people sleep during the night, different stages of sleep occur. These stages vary from light sleep to very deep sleep. These stages repeat over and over throughout the night and each stage plays an important role. Standard software packages exist today capable of scoring an entire night worth of sleep data to determine the different amounts of time spent in each stage.

These polysomnographs, or sleep studies, are extremely important to doctors who treat patients with sleep disorders. There are many types of sleep disorders, ranging from mild problems like insomnia and snoring, to sleep apneas and hypopneas that may be fatal. Thus, it is very important to use this specialized equipment to study the sleep patterns of these afflicted people.

Materials

- CleveLabs Kit
- CleveLabs Course Software
- A laptop computer (recommended, but if not available, a desktop computer in the same room where the subject will be sleeping)
- Nasal Cannula Thermocouple
- Respiratory Effort Belt
- MATLAB[®] or LabVIEWTM





Figure 1: The BioRadio can be used to perform an overnight PSG study. The signals displayed from top to bottom include two channels of EEG, two channels of EOG, chin EMG, ECG, breath rate from an oral/nasal thermistor, and respiratory effort indicated by a piezoelectric belt. The time scale shown is 10 seconds.

Background

Purpose of sleep

The exact purpose of sleep is still not entirely known by scientists. As mentioned previously, scientists once theorized that the brain shut down during sleep. However, we now know that is not true. There is some research that believes the sleeping process is very important for strengthening neuronal connections. For example, during one experiment, researchers taught subjects a new concept. These subjects were then divided into two groups. The researchers deprived one group of REM sleep and left the other group to sleep naturally. It was found that the group deprived of REM sleep could not remember the concept as well as those who had REM sleep. This illustrates that sleep may be important in helping the brain establish connections between neurons and as a result, help to re-enforce learned material. Additionally, sleep is a time where cells can regenerate and repair themselves. In an experiment with rats deprived of REM sleep, it was found that their lifetime was shortened to five weeks from the normal two years. Rats deprived of all forms of sleep showed signs of immune system dysfunction, low body temperature, and body sores after three weeks. Students in college may have noticed this effect also when spending many late nights studying, and finding themselves

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with a cold in the weeks afterwards. Therefore, sleep plays an important role in cell regeneration and immunity. In fact, when people are ill, the body forms very powerful sleep-inducing chemicals. During an immune response to infection, the immune system creates cytokines. These cytokines induce sleep, helping the body to conserve energy to combat the infection.

An active inhibitory process induces sleep. This was discovered by stimulation of certain portions of the brain. Stimulating these areas prevented sleep from occurring. Using this knowledge, scientists were able to disprove the passive theory of sleep. In the passive theory it was believed that a certain portion of the brain became fatigued after a certain number of wakeful hours. In this active inhibitory theory, scientists found that the raphe nuclei, located in the lower pons and medulla portions of the brain, send out inhibitory signals to the thalamus, neocortex, hypothalamus and limbic system. These signals actively induce sleep.

Chemicals in the blood have also been found to cause drowsiness. Agents that block serotonin have been found to cause insomnia. Therefore it is believed that serotonin may play a role in sleepiness. When a person has been awake for a long period of time the body begins producing certain peptides. It is believed that these peptides in the blood stream cause sleepiness. The body also receives cues from the natural environment about when to sleep. These are guided by the circadian rhythm of the body, also known as the biological clock. One important signal used by the biological clock is the amount of light seen by a person. The amount of light seen by a person controls the production of melatonin, the hormone that causes sleepiness. During periods of darkness, more melatonin is produced.

Types of sleep cycles

When a person sleeps, they pass through two types of sleep, non-REM and REM sleep. The non-REM sleep can be divided into four stages. In all, a person cycles between a total of 5 different stages of sleep during the night. The different stages of sleep can be determined by monitoring an EEG of the person while they sleep.

A person begins in stage 1 sleep, and then goes to stage 2, 3, and then 4. This process then reverses, and after stage 1 sleep, the person goes into REM sleep. The process is then repeated. Each sleep cycle (stages 1-4 and REM sleep) lasts from 90-100 minutes, and is repeated approximately 4-5 times during the night. Initially when the person is first asleep, more time is spent in deep sleep, stages 3-4. However, as the night goes on, the sleep cycle begins to shorten. Less time is spent in deep sleep, while more time is spent in REM sleep. Normally, about 45% of total sleep time is spent in stage 2 sleep, and 25% in REM sleep. The remaining 30% occurs in the other stages. As people age, the number of hours they sleep per night typically decreases and the percentage of time spent in stage 4 and REM also decreases.

Stage 1 sleep is characterized as the light sleep one experiences when just starting to fall asleep. The EEG recording shows alpha wave activity associated with quiet wakefulness. During stage 1 sleep, a person is easily woken, and may remember strange fragmented images and thoughts. The eyes move slowly and muscle activity slows. Also quite commonly noticed is hypnic

myoclonia, the sudden muscle contractions that occur when the body feels that it is starting to fall or slip.

Stage 2 sleep occurs after stage 1, and at this time, eye movement stops and the measured EEG shows slower brain activity in the theta band. This activity may be interrupted by brief bursts of waves in the alpha frequency called sleep spindles. Stage 3 sleep also shows theta wave activity, but fewer sleep spindle complexes.

In stage 4 sleep, the EEG slows even further to delta waves, a very low frequency signal. Stages 3 and 4 are considered deep sleep, and it is extremely difficult to wake a person in this stage of sleep. No eye movement or muscle activity is noted, and people who are awakened at this time of sleep will feel disoriented and confused. In deep sleep, the blood pressure and pulse decrease, blood vessels dilate, muscles become relaxed, and the body goes into a relaxed restful state.

REM sleep is one of the most interesting of the sleep stages. REM sleep is characterized by fast, rapid eye movements, giving this stage its name. These rapid eye movements are measured by the EOG during sleep. The person's breathing also becomes faster and shallower and the heart rate and blood pressure increases. Also, very importantly, muscle movement is inhibited during REM sleep by activating the pons in the brain, causing excitation of the medullary inhibitory area. The purposes of this are to inhibit motor neurons and paralyze muscle movement. The lateral motor strip also helps to inhibit muscle movement in the spinal column by active inhibition by the pons. REM sleep is the time that most people have dreams. Many of these dreams are strange and nonsensical images. Researchers believe that during REM sleep, the brain attempts to categorize and assimilate the information processed during that day, and a result of this attempt to organize random information are dreams. People typically spend two hours each night dreaming, however, they may not remember any of these dreams by the time they wake.

During REM sleep, not only are the eyes active, but the brain is active as well. The brain shows beta wave activity, very much like the beta wave activity that is recorded when a person is alert and awake. In fact, by looking at the EEG itself, one cannot distinguish between awake and alert activity from REM sleep. Thus, this process is called paradoxical sleep, since the EEG shows wakeful brain activity, even though the person is clearly asleep. People in REM sleep are more difficult to wake than people in stage 3-4 sleep, however, when people wake up in the mornings, they most likely wake up after a REM sleep stage. People who have been sleep deprived or extremely tired may not go into a REM sleep stage. Instead, more time is spent in deep sleep, such as stages 3-4, than in REM. Another indicator of REM sleep is the chin EMG measurements. As mentioned before, as a person enters REM sleep, the brain sends a signal to inactivate motor neurons during sleep, disabling muscle movement. Another consequence of this motor inactivation is decreased muscle tone. The chin EMG is used to detect this. As a person enters REM sleep, the chin EMG measurement will show muscle tone to be less than other stages of sleep. Hyperpolarizing motorneurons prevent most skeletal muscle activity during this stage.

As mentioned in the introduction, many tools that we have previously covered are used to determine when the body is in REM sleep. The EEG is used to find beta wave activity, the EOG

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is used to measure rapid eye movement, the ECG measures heart rate, EMG is used to measure decreased muscle tone, and respiratory effort and rate is determined by a respiratory belt and nasal cannula.

Sleeping Disorders

Sleeping disorders are the main reason why sleep studies are conducted. At least 40 million Americans suffer from different sleep disorders. These disorders can cause problems at work, social life, and may even endanger the person's life if left untreated. The most common types of sleep disorders are sleep apnea, insomnia, and narcolepsy. Narcolepsy and sleep apnea are the most life threatening.

Sleep apnea occurs when a person suddenly stops breathing during sleep. Frequently, people who snore loudly or are overweight are at risk for sleep apnea. However, not all people who snore are at risk for sleep apneas. People with sleep apnea suddenly stop breathing because the upper airway (Fig 2) collapses during the person's efforts to inhale air. While the person's brain becomes oxygen deprived, the brain responds by causing the person to gasp for air. As a result the person awakens and the sleep cycle is disrupted. Sleep apneas result in tiredness and irritable in the morning and can cause headaches due to several periods of oxygen deprivation. For some high-risk people, obstructive sleep apnea may cause death due to respiratory arrest during deep sleep. A sleep study can diagnose obstructive sleep apnea. An airflow sensor is used to monitor breathing during sleep. A clinician can diagnose apneas by reviewing the saved data from the sleep study.

These pauses in breathing during sleep can be classified into apneas and hypopneas. An apnea is defined as an 80% decrease or greater in breathing that lasts for more than ten seconds. Hypopneas refer to a pause in breathing that lasts more than ten seconds combined with a partial awakening or decrease in blood oxygen. Losing weight and changing ones sleeping habits by sleeping on the side or back can help to treat sleep apneas. Other methods include using a continuous positive air pressure (CPAP) machine. A CPAP supplies constant air pressure to the airway to keep it open during the night through a mask worn by the patient. However, CPAP machines are cumbersome to use and may cause irritation and dryness in the airways due to constantly moving air. In more extreme cases, patients may elect to undergo surgery to correct apneas.



Figure 2: Respiratory signals are very important to monitor during sleep studies to detect apneas.

Insomnia is experienced by nearly all people at some point in life, however, some people have extreme cases of insomnia that require medical attention. Normally, mild insomnia is caused by stress, diet, jet lag and other factors, but serious insomnia may be an indication of a deeper medical problem. It is very important to have these serious cases of insomnia diagnosed, since prolonged insomnia leads to immune system weakness, and may eventually cause death indirectly. The most common treatment for insomnia is sleeping pills, however, long-term use of these pills may actually cause addiction and cause serious problems with good natural sleep.

Another common sleep disorder is narcolepsy. Narcolepsy is when a

person feels the urge to suddenly take naps during the day, even when they have had a normal amount of sleep the night prior. These narcoleptic attacks can last between several seconds to 30 minutes, and while in a narcoleptic attack, these people experience hallucinations and may be in a catatonic state, suggesting that REM sleep is occurring at these points. The cause of narcolepsy is often found to be neurological disease or injury to the brain. Drugs can control narcolepsy. Narcolepsy is extremely dangerous, since a person may have a narcoleptic attack during a crucial time like driving a car or operating heavy machinery.

Pulse oximeters are also used for sleep studies. Pulse oximeters measure the percent saturation of oxygen in the blood stream. If a person has an apnea during sleep, the oxygen saturation in the blood will drop since the person is not breathing. A pulse oximeter is not used in this laboratory. A thermistor and respiratory effort belt are used. If an obstructive apnea were to occur, one would notice a flat line on the thermocouple graph even though the respiratory effort belt indicates the person attempting to breathe.

Experimental Methods

Experimental Setup

This laboratory will record eight channels of standard PSG signals including 2 channels of EEG, 2 channels of EOG, 1 channel of ECG, 1 channel of EMG, 1 channel of airflow, and one channel of respiration.



- 1. As you may have noticed, the sleep experiment will involve many concepts and techniques that have been discussed in previous labs. In fact, you may want to review these laboratories as it may help the setup run smoother. Proper electrode and sensor setup is a very important part in a sleep lab. If any of the electrodes fall off during the course of the night, the experiment may need to be repeated. NOTE: This laboratory requires that the data collection computer have approximately 100MB of free disk space to save the data. Unlike previous labs, you will be recording biopotentials from all eight channels for approximately eight hours. Therefore, you will be handling significantly larger amounts of data. In addition, you MUST install a new set of batteries into the BioRadio unit before the start of this study.
- 2. During the polysomnograph you will measure two channels of EEG, two channels of EOG, EMG from the chin, ECG, respiratory effort, and oral/nasal airflow. It is very important that you properly prepare the electrode sites before applying the electrodes.
- 3. One gold cup electrode should be placed on each of the mastoids of the subject (behind the ears as in the EEG labs). These two electrodes will be used as references. Therefore, you should connect the output of one of these electrodes to the -1 input and the other to the -2 input on the BioRadio.
- 4. We also need a ground electrode. Place a gold cup electrode at position FPz. Connect the other end of the gold cup electrode to the "GND" input of the BioRadio.
- 5. Next, we will attach the gold cup electrodes to the back of the subject's head to monitor EEG. EEG will be recorded from sites O1 and O2. You should connect these electrodes to inputs +1 and +2 on the BioRadio.
- 6. You may want to wrap the head with gauze after these electrodes are in place (Fig 3). It is very important that these electrodes stay attached throughout the course of the night. The subject will be rolling their head around on a pillow throughout the course of the night and if the head is not wrapped this may pull the electrodes off of the head. The gauze can be used to wrap a headband around the head. The headband should wrap around the front of the head in order to cover the reference and ground electrodes and then around the back of the head in order to cover the two EEG electrodes. The headband wrap should be taught to hold the electrodes in place, but not so tight that it is uncomfortable for the subject. It should be wrapped in such a way that it holds the electrodes against the head, but does not pull them toward the top of the head.
- 7. Next we will place the EOG electrodes on the subject. We will use cloth snap electrodes to measure EOG. Two channels of EOG will be recorded. Properly prepare the surface of the skin before attaching the surface electrodes for EOG. Attach a snap electrode just left of the subject's left eye, and slightly above the midline of the eyes on the left temple. Attach another electrode just right of the right eye, but slightly below the midline of the Using a snap lead, connect the snap electrode near the eyes on the right temple. © 2006 Cleveland Medical Devices Inc., Cleveland, OH.

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subject's right eye to input +3 on the BioRadio. Using a snap lead, connect the snap electrode near the subject's left eye to input +4 on the BioRadio.

- 8. Unlike in the previous EOG sessions, we will now only be recording the AC component of the EOG signal. Therefore, we will use the same references as we are for the EEG recordings. Therefore, using a jumper, connect -1 to -3 on the BioRadio unit. Using another jumper, connect -2 to -4 on the BioRadio unit.
- 9. Chin EMG will also be recorded for this polysomnograph study. Two snap electrodes will be placed on the chin approximately one inch apart. It is important that you properly prepare the electrode sites before placing the electrodes on the chin. Place the two snap electrodes on the chin. Using snap leads, connect one of the snap electrodes to input +5 on the BioRadio and the other to input -5.
- 10. Snap electrodes will also be used to measure ECG. It is important that you properly prepare the electrode sites before applying the snap electrodes. One snap electrode should be placed on the skin above the right clavicle and one should be placed on the skin above the left clavicle. Using a snap lead, connect the electrode above the left clavicle to input +6 on the BioRadio unit. Using a snap lead, connect the electrode above the right clavicle to input -6 on the BioRadio unit.
- 11. The oral/nasal thermocouple will be used to monitor airflow from the subject. The thermistor should be mounted between the nostrils and upper lip of the subject (Figure 3). There are three prongs on the thermocouple. The side with two prongs should point up, one prong slightly into each nostril. The prong on the bottom of the thermocouple should

be bent around so that the tip is positioned directly in front of, but not in, the subject's mouth. The thermistor can be taped in place and the wires can be run over the ears and over the back of the head. Two other pieces of medical tape can be placed over the wires on the face to further hold the thermistor in place and keep the leads from dangling around on the face. Connect one of the leads from the thermocouple into the +7 input on the BioRadio and the other into the -7 input.

12. Finally, the provided piezoelectric respiratory effort belt should be placed around the torso of the subject. Securely fasten the respiratory effort belt around the subject just above the stomach and below the rib cage. In this position both



Figure 3: Wrapping the head with gauze during a sleep study helps to ensure the EEG, ground, and reference electrodes stay in place. In addition, the wires should be run around the back of the head for comfort.

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diaphragm and chest breathing should be captured. Connect the leads of the respiratory effort belt to the inputs of channel 8 on the BioRadio unit.

- 13. To avoid tangling and discomfort for the subject, the lead wires from the thermistor, the ground and reference electrodes, chin EMG, and ECG electrodes should be run around the back of the head. It is also a good idea to place a piece of medical tape over the EOG, EMG, and ECG snap electrodes to ensure they stay attached throughout the night. The BioRadio unit can be worn on the included armband during the night.
- 14. Turn the BioRadio unit on.
- 15. Check that you have properly hooked up the system using the CleveLabs software. Log in and click on the "Polysomnography" laboratory session to begin. Your BioRadio should be programmed to the "LabSleep" configuration.
- 16. Click on the green start button to begin scrolling real time data.
- 17. You may need to adjust the amplitude and time scales. Verify both channels of EEG by instructing the subject to close their eyes and relax and check for alpha waves. Verify the ECG signal is properly being recorded based on your ECG experience. Verify the EOG is being properly recorded by having the subject move their eyes to the left and right. Verify that the chin EMG is working by having the subject scrunch their chin. Verify the thermocouple and respiratory belt are working by having the subject take normal and deep breathes.
- 18. Turn the BioRadio off.

Procedure and Data Collection

- 1. Just before the subject goes to sleep for the night, they should turn on the BioRadio unit and also start Polysomnography Lab software. Your BioRadio should be programmed to the "LabSleep" configuration. They may use a laptop computer for this experiment or they may use a desktop computer that is located near where they sleep.
- 2. First, they should once again check to make sure that all of the signals are being displayed properly. They should then begin saving data to a file named "sleep".
- 3. Then they should leave the software running and transmitter on and go to sleep.
- 4. In the morning, when the subject wakes up they should stop the software from running and turn off the BioRadio unit. They can also remove all electrodes and sensors.



Data Analysis

- 1. Using the post processing toolbox, MATLAB, or LabVIEW, open the data file "sleep" that you created.
- 2. Try to find different stages of sleep according to what you know about sleep stages and signal processing. Capture a screen shot of each section that you select. Be sure to examine at least a few sections of data from each hour of the sleep study. Do you see the sleep cycle repeating?
- 3. For each of the different sections of data that you annotated, eyeball and note the average breathing rate, heart rate, EEG and EOG frequencies, and EMG amplitude. Create a graph that plots these average values against the different sleep stages. What do you see happening in the different stages? Does this make sense? Explain.

Discussion Questions

- 1. Overnight sleep studies can be used to determine if a person has an obstructive sleep apnea. Think about the signals were recorded. Which would be important in diagnosing an obstructive sleep apnea? Sketch a plot of these signals to illustrate normal sleep and an apnea.
- 2. There is a sleep disorder called REM sleep behavior disorder in which the individual's body moves according to what he/she is dreaming. For instance, when a person suffering from REM sleep behavior disorder dreams of throwing a Frisbee, they may find that their arm hurts in the morning because they have slammed it into the wall while asleep. Explain what mechanisms are (or are not) involved in this individual, and suggest what may help to reduce the severity of the disorder.
- 3. Why is it important to be aware the file size for sleep studies? What can be done to keep the file size as small as possible but without sacrificing quality? And what quality is being sacrificed in order to keep the file size small?
- 4. Why is the EEG, during REM sleep, considered paradoxical beta? What mechanisms ensure that it is paradoxical?
- 5. Explain any artifacts that you noticed in the data from the sleep study and what their sources may be.
- 6. Instead of simply eyeballing the data, we are now going to make actual calculations using MATLAB or LabVIEW. Create a software program that will calculate the actual peak amplitudes and frequencies over each minute of your entire saved data file. Now plot these values over the course of the night and examine if you see any trends or cycles occurring.

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7. Suggest how an automated program that could be developed to automatically score sleep studies could work.



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