

INTRODUCTION

"Welfare" can be described as a dynamic balance between an animal and its environment, encompassing both its internal and external conditions.

The animal must often make efforts to maintain or restore this balance, which can sometimes lead to physical and mental suffering that may harm its health and productivity.

The welfare of farm animals is assessed using four types of complementary and essential measures: zootechnical, semiological, physiological, and ethological measures.

The latter present numerous advantages and are increasingly popular, as they allow study of animal adaptation, behavioral disorders, as well as their motivations and preferences.

In practical work sessions, students must participate in different activities, so that they can verify their ability to mobilize all the theoretical concepts acquired, enabling them to contribute to animal benevolence and good treatment.

Students will be assessed at the end of each activity through tests and a practical exam.

General objectives

The practical work in ethology and animal welfare aims to:

- Define stress in animals;
- Identify types of stress in animals;
- Differentiate between stress factors;
- Verify the animal's response to stress;
- Assess the animal's response to stress.

PRACTICAL WORK 1: STRESS TEST AND RESPONSE ASSESSMENT IN SHEEP FARMING

SPECIFIC OBJECTIVES

At the end of this chapter, you will be able to:

- List animal welfare indicators in sheep farming.
- Identify types of stress in sheep.
- Differentiate between stress factors in sheep farming.
- Verify the sheep's response to stress.
- Assess the animal's response to stress in sheep farming.

1. REVIEW OF ANIMAL WELFARE AND THE FIVE (5) FREEDOMS (FUNDAMENTAL NEEDS OF THE ANIMAL)

An animal's welfare is the positive mental and physical state related to the satisfaction of its physiological and behavioral needs, as well as its expectations.

- Welfare is considered as the absence of physical and mental suffering.
- This state varies according to the animal's perception of the situation.
- Animal welfare is based on 5 freedoms:
- Freedom from hunger and thirst: physiological freedom;
- Freedom from discomfort: environmental freedom;
- Freedom from pain and disease: health freedom;
- Freedom to express normal behavior: behavioral freedom;
- Freedom from fear and distress: psychological freedom.

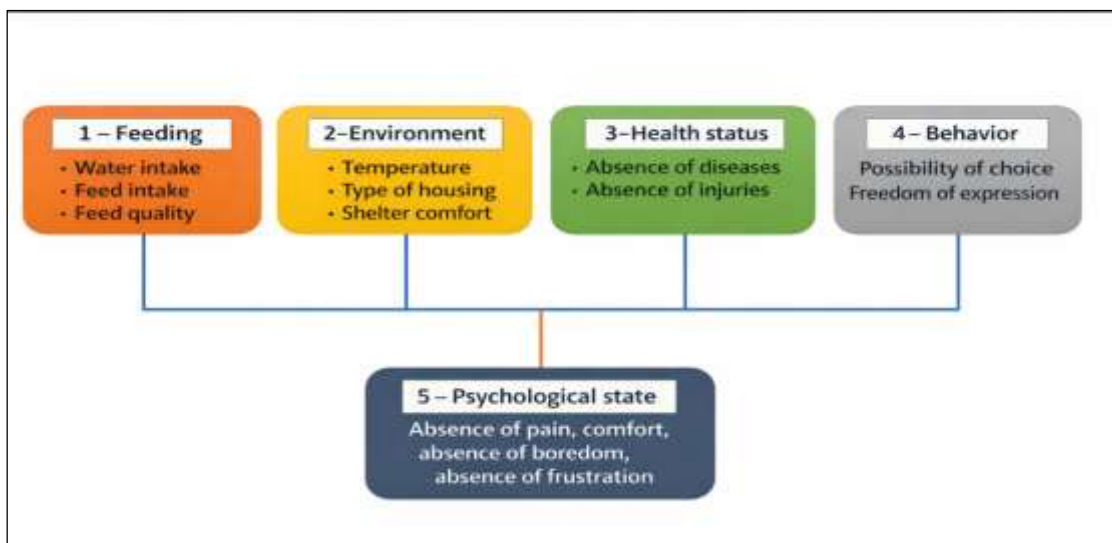


Figure 1: The five domains of action for animal welfare

✓ In Figure 1, each of the four domains—nutrition, environment, health, and behavior—will influence the fifth domain, the animal's psychological state.

Example

- If the animal is deprived of food or water (domain 1), this will result in the sensation of hunger and thirst (domain 5).

2. INDICATORS FOR ASSESSING ANIMAL WELFARE IN SHEEP FARMING

2.1. MEASURES CARRIED OUT ON THE ANIMAL'S ENVIRONMENT

These are input-based indicators:

- Number of waterers,
- Number of feeders,
- Characteristics of pens (area, ventilation, temperature, bedding...),
- Farming practices (transport, restraint, care, feeding...)
- Recording building measurements (accessibility of troughs/water points, cleanliness...)
- Asking the farmer about their farming practices (feeding, bedding, care...)

These indicators are easy to measure.

2.2. MEASURES CARRIED OUT ON THE ANIMAL

These are direct output-based indicators at the animal level. They are considered experimental (require time and expertise).

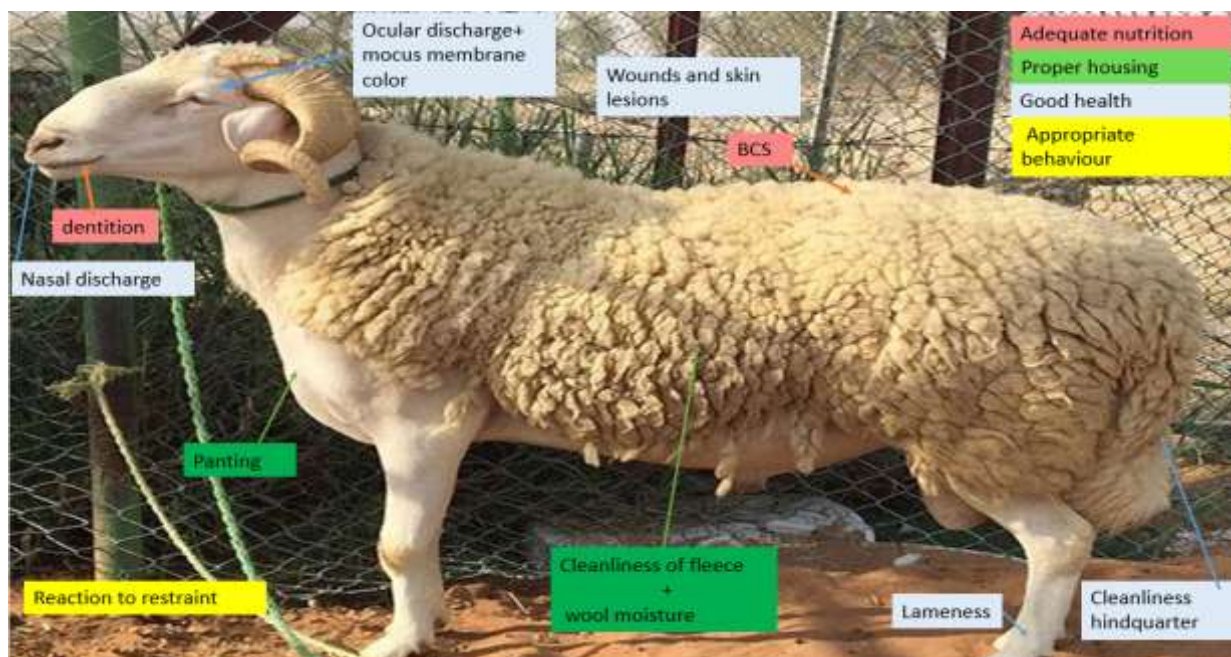


Figure 2: Individual measurements on sheep

2.3. PERFORMANCE MEASURES

These are indirect output-based indicators on the animal, at the herd level.

Examples

- Mortality rate,
- Production rate (milk production, weight gain, prolificacy),
- Fertility rate,
- Dystocia rate.

3. STRESS AND TYPES OF STRESS

3.1. DEFINITION OF STRESS

Stress is the set of reactions of an organism subjected to constraints caused by external aggressions related to its environment.

- It constitutes the state of an animal that is subjected to conditions that may have unfavorable consequences on it; and which force it to react.
- Stress can have harmful consequences on the animal's health and on its zootechnical performance.

3.2. TYPES OF STRESS

3.2.1. Acute stress

Which depends on the punctual variation of a stressor, of limited duration in time; that is to say, stress diminishes rapidly with the disappearance of the cause.

Examples

- Rapid drop or increase in temperature,
- Sudden noise,
- Unexpected appearance of a predator.

3.2.2. Chronic stress

When the action of the stressor on the animal's environment lasts a long time or is repeated.

- In a stressful situation that lasts over time, a pathology can occur and sometimes cause the animal's death.

Examples

- Repeated attacks from a dominant conspecific,
- Chronic thermal stress,

- Lack of food,
- Long-duration transport.

4. MAIN STRESS ACTORS

Faced with a stressful situation, the animal organism activates two complementary systems: a rapid response (acute stress) via the sympathetic nervous system and a slower response (chronic stress) via the hypothalamic-pituitary-adrenal axis (HPA axis).

4.1. THE CENTRAL NERVOUS SYSTEM: The prefrontal cortex plays a role in cognition (action planning), and the limbic system (including the amygdalae: the brain's fear center) play a role in emotion regulation.

4.2. THE AUTONOMIC NERVOUS SYSTEM: As soon as the stress factor is perceived, the sympathetic nervous system is activated and triggers the release of adrenaline and noradrenaline (catecholamines) by the adrenal medulla.

4.3. THE HYPOTHALAMIC-PITUITARY-ADRENAL AXIS (HPA): The HPA axis involves three main actors: the hypothalamus secretes CRF (Corticotropin Releasing Factor), which activates the pituitary gland, which releases ACTH (Adrenal Corticotropic Hormone) into the blood. ACTH then stimulates the synthesis and release of cortisol (glucocorticoids) by the adrenal glands.

4.4. THE IMMUNE SYSTEM: Release of pro-inflammatory cytokines during acute stress to activate immune defenses. In chronic stress, cortisol causes immunosuppression, increasing susceptibility to infections.

Stress hormones such as adrenaline and cortisol are responsible for the animal's reaction during stress.

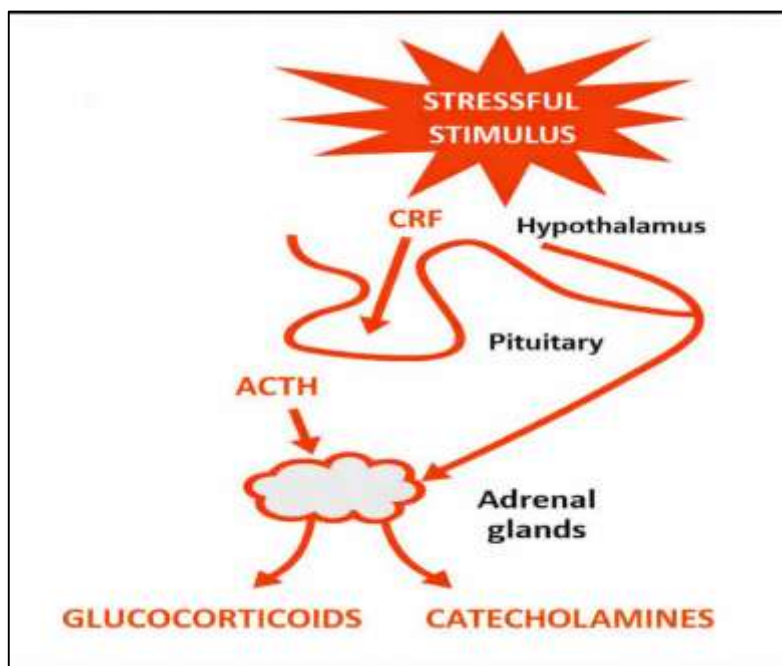


Figure 3: Main stress actors

5. STRESS FACTORS

The causes of stress are multiple; they can be related to environmental factors, social factors, poor farming practices, and various pathologies (Table 1).

Table 1: The different stress factors in sheep farming

Environmental Factors	Social Factors	Factors Related to Farming Practices	Factors Related to Pathologies
Temperature variation (heat, cold, rain, wind)	Grouping	Feed (insufficient, excessive, unbalanced) Lack of water Unsuitable water	Pathogenic organisms: Parasites, bacteria, viruses
Humidity	Isolation (separation of the animal from its conspecifics)	Housing conditions Surface area relative to number of animals	Pain related to trauma
Ventilation	Density (number of animals)	Animal handling	Injuries (fractures...)
Luminosity	Weaning (separation of young from mother)	Transport	Metabolic disorders
Change of environment	Relationship with other animals (attacks by a dominant conspecific, predation)	Relationship with humans (approach, handling, various treatments, such as shearing)	-

6. ANIMAL ORGANISM'S RESPONSE TO STRESS

6.1. ACUTE STRESS

✓ Faced with an acute stress situation, the animal responds by:

6.1.1. Behavioral changes: The animal will react by:

- Either defending itself,
- Either fleeing during predation, aggression, or handling,
- Or facing the situation (enduring).

6.1.2. Physiological changes: The organism responds with a discharge of adrenaline (stress hormone) which causes:

- Increased heart rate (tachycardia),
- Increased respiratory rate (tachypnea),
- Increased body temperature (hyperthermia),
- Increased blood glucose (hyperglycemia).

Other signs:

- Slowing of digestion,
- Pupil dilation,
- Sweating (activation of sweat glands).

Note

- This adrenaline discharge stops with the disappearance of the unfavorable situation.
- In acute stress, the organism's response is short-term, rapid, and strong.

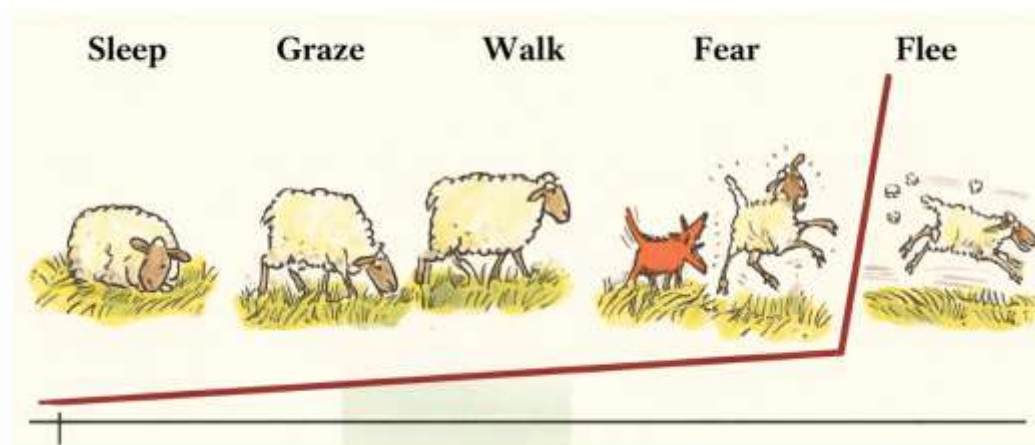


Figure 4: Animal's response to acute stress (presence of a predator)

Humans and dogs (stress factors) are seen by sheep as a threat. Their intervention generally causes a flight response from the animals (they try to jump over barriers, press themselves against them).

6.2. CHRONIC STRESS

During chronic stress, for example: poor housing conditions, prolonged malnutrition, persistent bad climate, presence of pathologies:

- The situation persists over time.
- The reactions of the autonomic nervous system and endocrine system follow one another with hormonal secretions (such as cortisol), leading to metabolic and biochemical modifications.

Note

- Cortisol is secreted to help adrenaline maintain a high energy level.
- Its main role is to increase blood glucose levels to provide necessary energy to muscles, brain, and heart.

✓ After a more or less long time (a few days, weeks, months, years):

- Either the unfavorable situation disappears (change of season, improvement of farming conditions, healing...). Cortisol also constitutes a message to stop the production of hormones that trigger the HPA axis cascade. It activates its receptors in the brain (hypothalamus and pituitary gland) and this inhibits its own secretion. This is a negative feedback loop, because the quantity of the product negatively affects its own production. It thus allows a return to homeostasis after the disappearance of the stress factor.
- Or the animal adapts to the situation (for example adaptation to changes in farming techniques, to environmental change). (See subsection 6.3. Adaptive capacities).
- Or on the contrary, it fails to adapt, and loses its physical and psychological capacities. The hyperactivity of the HPA axis in response to chronic stress is driven by hypersecretion of CRH at the hypothalamic level and may be accompanied by a deficiency in the axis feedback. This phase appears when regulatory systems are overwhelmed and immune function is affected, with the appearance of pathologies that can lead to the animal's death.

Note

- Most of the time, conditions disappear or are quickly eliminated, or the animal adapts to them.
- One can even say that the regular appearance of stressful factors is necessary for the animal, because it allows it to test and maintain its reaction and adaptation capacities.



Figure 5: Animal's response to acute and chronic stress

6.3. ADAPTIVE CAPACITIES

- The adaptive capacities of a farm animal characterize its abilities (capacity) to adapt to its environment, in response to farming conditions.
- This capacity depends on the animal's genetics, its experience, and its learning.
- Then, adaptation processes (coping) occur, comprising neuro-hormonal and physiological mechanisms as well as behavioral adjustments.

✓ In case of success, homeostasis leads to welfare.

✓ In case of adaptation failure, this leads to stress, increased susceptibility to pathology, suffering, and poor welfare.

- Indeed, physiological and behavioral adaptation processes are triggered when the situation is perceived as a threat by the animal.
- In conclusion, the stress response depends on how the animal evaluates the situation.

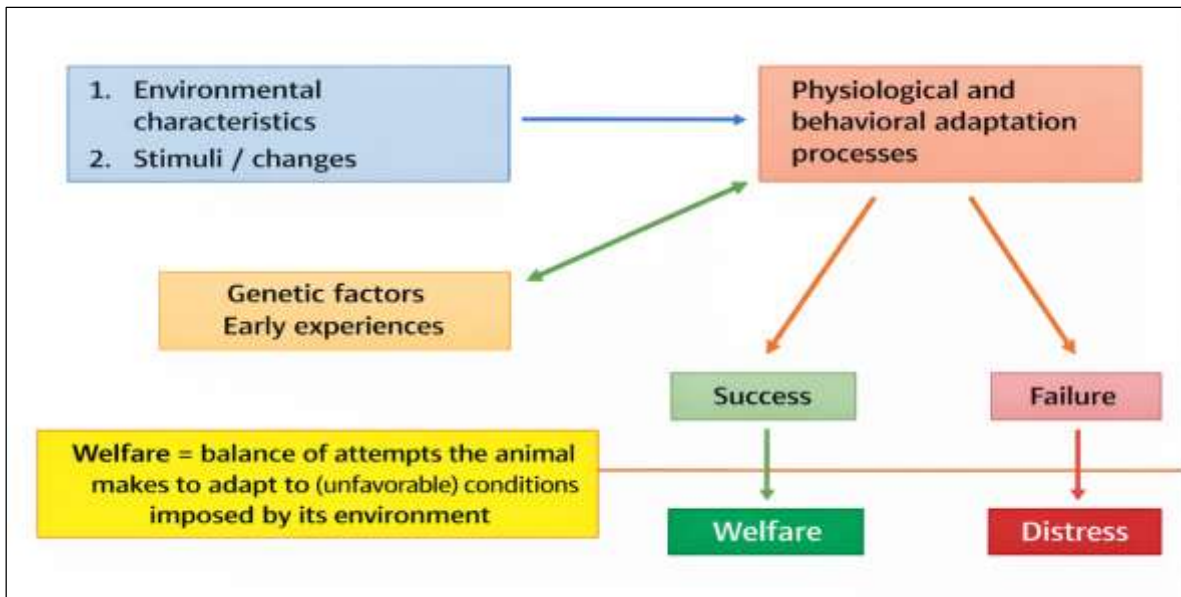


Figure 6 : Coping concept

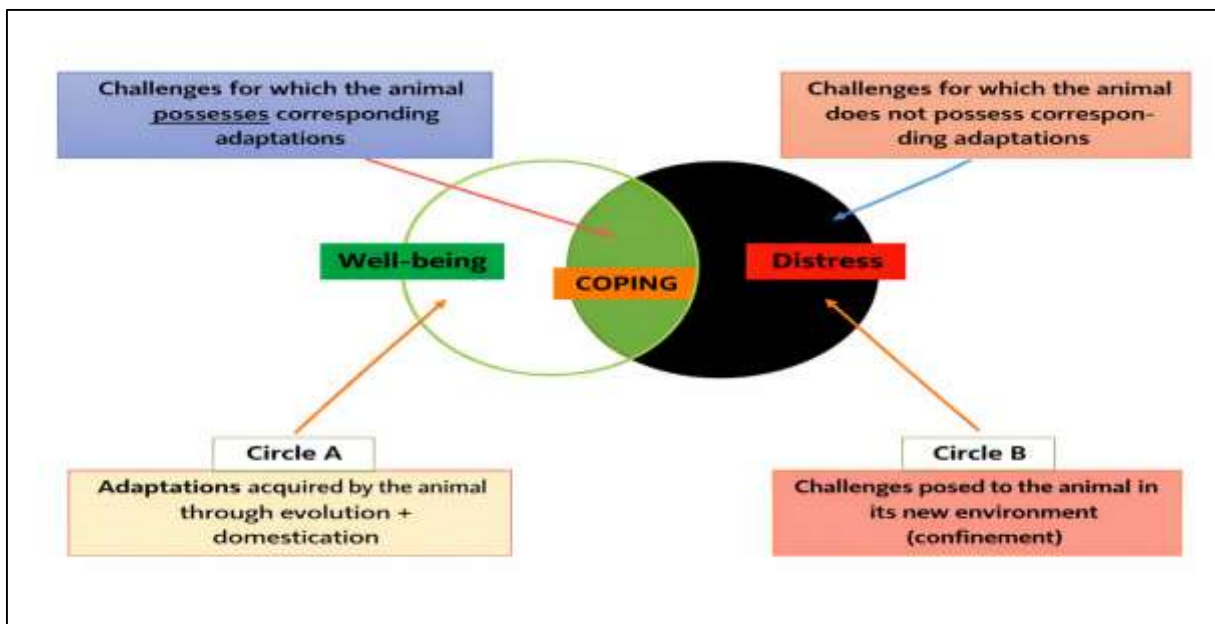


Figure 7: Model of animal adaptation to its environment and animal welfare assessment

In Figure 7, the circle on the left represents the animal's characteristics and the circle on the right represents the environment.

The more the circles overlap, the easier the animal's adaptation and the greater the welfare.

When the two circles no longer correspond at all, the environment can no longer satisfy the animal's needs, which impacts its health and emotional state. This leads to poor welfare in the animal.

7. ASSESSMENT OF STRESS RESPONSE

7.1. BEHAVIORAL CHANGES

- Behavior constitutes an early indicator of animal discomfort.
- One can observe a modification of the animal's behavior (flight, fear, aggressiveness, isolation, underfeeding...).

Example

Fear is a highly developed emotion in sheep; it varies according to numerous factors specific to the animal (breed, sex, age, physiological state) or from the environment (farming method, frequency and nature of contact with humans, environment, dog attacks...).

7.2. PHYSIOLOGICAL CHANGES

When measuring physiological changes, one can observe:

- An acceleration of heart rate (tachycardia),
- An acceleration of respiratory rate (tachypnea),
- An increase in blood concentration of stress hormones (adrenaline, cortisol), glucose...

Note

- The changes brought to the animal organism are the consequence of stress hormone secretion by the adrenal glands (hypothalamic-pituitary-adrenal axis).
- The best known is the increase in heart rate.
- In sheep, analyses show that compared to a normal rhythm of 75 beats per minute, the increase ranges from 20% during isolation to 84% when approached by a person accompanied by a dog.
- The increase in blood cortisol concentration is another known consequence.
- Compared to a baseline level of 3.6 pg/100 ml of plasma, concentrations can approach or even exceed 10 pg/100 ml in case of dog attacks.

7.3. ZOOTECHNICAL PERFORMANCE

The stress response induces different changes that can have negative effects on zootechnical performance.

- These effects include among others changes in immune function and consequent increased susceptibility to diseases, reduction in feed intake and rumination.
- If significant stressful conditions persist, they cause a reduction in reproductive performance (fertility and prolificacy), and growth delays in fattening animals.

7.4. PATHOLOGICAL DISORDERS

- Stress has an effect on the immune system and leads to immunosuppression.
- If conditions are truly unfavorable, pathological disorders can occur.

Example

- In case of uncontrolled farming management (poor feeding, poor hygiene, or poor ventilation),
- When several stressful factors accumulate during transport (temperature, noise, vehicle movement, and absence of food and water...), respiratory diseases may appear.

8. STRESS MANAGEMENT

Respect for animal welfare and good control of farming practices help mitigate the effects of stress on the animal.

Example

- Provision of fresh water and balanced food,
- By ensuring good health and avoiding suffering,
- By providing an appropriate environment (sufficient space, shelters, comfortable rest area, contact with animals, etc.).
- The animal's good health (absence of lesions, injuries, diseases, etc.) and zootechnical performance (growth, fertility, productivity, etc.) can testify to the animal's welfare state.
- Health monitoring of farms by veterinarians contributes to animal protection and their health protection: treatments against parasites, regularly performed vaccinations, lameness treatments, etc.

9. WORK PROTOCOL

9.1. WORKPLACE: at the farm / sheep farm level

9.2. NECESSARY EQUIPMENT: surgical alcohol, cotton, electronic thermometer, stethoscope, syringe, glucometer.

9.3. WORK IMPLEMENTATION: expose sheep to acute stress (loud noise, animal restraint, presence of strangers).

9.4. ASSESS THE ANIMAL'S RESPONSE TO STRESS:

Identify behavioral changes in sheep such as flight, aggressiveness, and fear, according to the animal's perception of the situation.

Also measure physiological changes by:

9.4.1. Non-invasive method

A. Necessary equipment: electronic thermometer and stethoscope.

B. Temperature measurement: in sheep, temperature is taken rectally by introducing an electronic thermometer into the anal orifice so as to touch the rectal mucosa. Physiological norms for temperature in an adult sheep are between 38.5 and 39.5°C.

C. Heart rate measurement: in sheep, a stethoscope can be used to measure the number of heartbeats.

To do this, the stethoscope is placed under the tip of the elbow, that is, between the humerus and the rib cage.

Heart rate can be measured over a period of 30 to 60 seconds.

Physiological norms in adult sheep are between 75 and 85 beats per minute, while in young sheep, it varies between 90 and 100 beats per minute.

D. Respiratory rate measurement: in sheep, respiratory rate is measured by counting the number of inspiration and expiration movements observed at the flank hollow level, mainly on the right side, over a duration of one minute.

Physiological norms for respiratory rate in adult sheep are between 12 and 15 movements per minute, while in young sheep, it is between 15 and 18 movements per minute.

It is essential to compare the obtained results with these physiological norms.

9.4.2. Invasive method

A. Necessary equipment: alcohol, cotton, syringe, glucometer.

B. Blood sampling: blood sampling in sheep is performed from the jugular vein, located at the animal's neck level.

To locate the vein, pressure is applied downstream. Before sampling, it is essential to disinfect the sampling area using an antiseptic, such as alcohol.

Then, the syringe needle is placed at a 45° angle and carefully introduced into the vein. Once the needle is in place, blood is gently aspirated.

Blood glucose in sheep is: 0.3 - 0.6 g/l.



Figure 8: Rectal temperature measurement (personal photograph)



Figure 9: Heart rate measurement (personal photograph)



Figures 10: Blood sampling in sheep from the jugular vein (personal photographs)

PRACTICAL WORK 2: STRESS TEST AND RESPONSE ASSESSMENT IN POULTRY FARMING

SPECIFIC OBJECTIVES

At the end of this practical work, you will be able to:

- Know the effect of temperature on poultry.
- Identify thermal stress factors in poultry farming.
- Determine the phases of thermal stress in poultry farming.
- Verify poultry response to thermal stress.
- Assess the animal's response to stress in poultry farming.

1. EFFECT OF TEMPERATURE ON POULTRY

1.1. BODY TEMPERATURE OF POULTRY

Poultry belong to the group of homeothermic animals capable of maintaining a constant internal body temperature (41.5°C for adults) for optimal functioning of vital organs.

✓ It is important that heat losses are identical to poultry heat production: this is called thermal balance. This mechanism is called thermoregulation.

- During the feather development phase (1 day to 3 weeks of age), they are sensitive to cold thermal stress.
- After this phase, which will only be complete from the 5th week of age, they present excellent insulation and will be rather sensitive to excess heat.
- Therefore, any thermal discomfort can have repercussions on the animal's physiological balance, its health status, and its zootechnical performance.

1.2. REGULATION OF BODY TEMPERATURE

The means of combating heat implemented by the organism are represented by the decrease in thermogenesis (decrease in heat production) and the increase in thermolysis (heat loss process).

✓ Heat elimination occurs through:

- Evaporation: through panting respiration.
- Conduction (contact of feet, breast with litter and walls),
- Convection (toward air and through feathers)
- Radiation (through air toward colder walls or litter).



Figure 11: Main modes of heat transfer between the animal and the environment

1.3. EFFECT OF FOOD INTAKE ON BODY TEMPERATURE

- Hens constantly adapt their feed intake according to ambient temperature.
- Food digestion represents the main mechanism for hens to generate metabolic heat.
- When exposed to colder temperatures, poultry normally ingest a greater quantity of food in order to increase and maintain their body heat.
- Consequently, as ambient temperature rises, hens tend to reduce their food consumption, which helps limit the increase in their body heat related to digestion.
- Fast-growing broiler chickens and high-production laying hens are particularly vulnerable to environmental challenges such as thermal stress, caused by high temperatures and significant humidity.
- This phenomenon results from their high nutritional needs, as food ingestion, digestion, absorption, and metabolism generate heat in their organism. In case of high ambient temperatures, this process can lead to thermal stress.

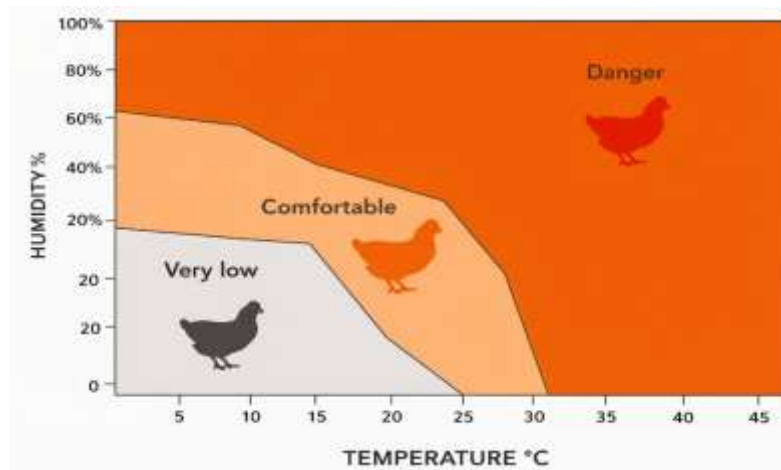


Figure 12: Influence of temperature and humidity rate on poultry comfort

✓ The illustration above indicates that a relative humidity between 45 and 75%, combined with a temperature below 30 degrees Celsius, remains favorable for poultry welfare.

Note

- Poultry do not have sweat glands and do not perspire. They evacuate body heat by panting (breathing with open beak) and by conduction (contact of plantar pads and the sternal part of the breast with litter).

2. THERMAL STRESS

This is an accumulation of heat in an organism, which prevents it from maintaining a normal body temperature.

- The notion of heat or exposure to high ambient temperature covers two different aspects: acute thermal stress and chronic thermal stress.

2.1. ACUTE THERMAL STRESS

Heat stroke, which is an acute thermal stress with very high temperature (exceeds 35°C) for a relatively brief time.

- This type of stress induces immediate reactions that converge toward a single objective: the animal's survival.
- Its main consequence is an increase in mortality, often by suffocation.

2.2. CHRONIC THERMAL STRESS

This type of stress appears during exposure to high ambient temperatures, generally of a cyclical nature (between 29 and 35°C during the day, cooler ambient temperatures during the night) and extending over relatively long periods, ranging from a few days to several weeks.

- In this type of exposure, mortality is only very slightly increased while zootechnical performance is largely affected.

3. CONTRIBUTING FACTORS

✓ Several factors favor the appearance of thermal stress.

3.1. AGE OF POULTRY

- The older the animal, the more sensitive it is to heat thermal stress.

3.2. AIR FLOW (VENTILATION)

- When ventilation is good, the temperature felt by the animal is lower.

3.3. POULTRY BEHAVIOR

- Hens/chickens that tend to huddle together find themselves in a thermal stress situation.

3.4. GROUP HOUSING SYSTEM

- Ventilation in group housing is not comparable to that of an open aviary system.

3.5. ACTUAL TEMPERATURE AND RELATIVE HUMIDITY IN THE BUILDING

- They are always different (often higher) than the outside temperature and humidity.

4. RESPONSES TO THERMAL STRESS

4.1. ACUTE THERMAL STRESS

✓ The indicative signs (behavioral and physiological changes) of decreased welfare in laying hens and broiler chickens during heat stress are:

- Wing spreading;
- Panting with open beak;
- Reduction in physical activity;
- Increase in water consumption;
- Decrease in food consumption.

Leading to:

- Increase in respiratory rate (tachypnea);
- Increase in heart rate (tachycardia);
- Increase in body temperature.



Figures 13: Turkey panting following increase in ambient temperature (personal photograph)

4.2. CHRONIC THERMAL STRESS

In laying hens, heat stress results in a reduction of available volumes of nutrients and energy, necessary for egg production.

✓ Clinical signs of heat stress in laying hens can also be:

- Increase in the number of thin-shelled eggs,
- Decline in egg production,
- Decrease in egg size.

✓ Heat stress can damage the intestinal wall and induce leaky gut syndrome.

✓ This syndrome manifests as:

- Loss of intestinal cell integrity: poultry are more sensitive to Salmonella infections and other diseases,
- Appearance of secondary infections if sanitary conditions are of low level,
- Reduction in nutrient digestibility level,
- High levels of endotoxins in plasma,
- Due to inflammation of intestinal villi, harmful microorganisms penetrate more easily into the organism and bloodstream.

5. THE FOUR PHASES OF THERMAL STRESS

5.1. AMBIENT TEMPERATURE ABOVE 25°C:

- Maintaining the hen's body temperature remains effective, staying at 41.5°C.

5.2. AMBIENT TEMPERATURE BETWEEN 25°C AND 30°C:

- The hen's body temperature increases by about one degree, reaching 42.5°C.
- To counter this, the hen accelerates its heart and respiratory rates.
- Evaporation through panting becomes the most important mode of heat elimination. The higher the ambient temperature rises, the more heat loss through panting becomes the sole fighting mechanism.

5.3. EXCESSIVELY HIGH AMBIENT TEMPERATURE :

- The hen can no longer resist, lies down, and its body temperature can reach 44°C.
- Its heart and respiratory rates increase significantly (200 movements/min), leading to dehydration.

5.4. IN CASE OF EXTREMELY HIGH AMBIENT TEMPERATURE:

- The hen enters vital distress, presenting a decrease in respiratory rate. If relative humidity is too high, evaporation becomes insufficient.
- Body temperature can then increase up to 47°C, leading to the animal's death.

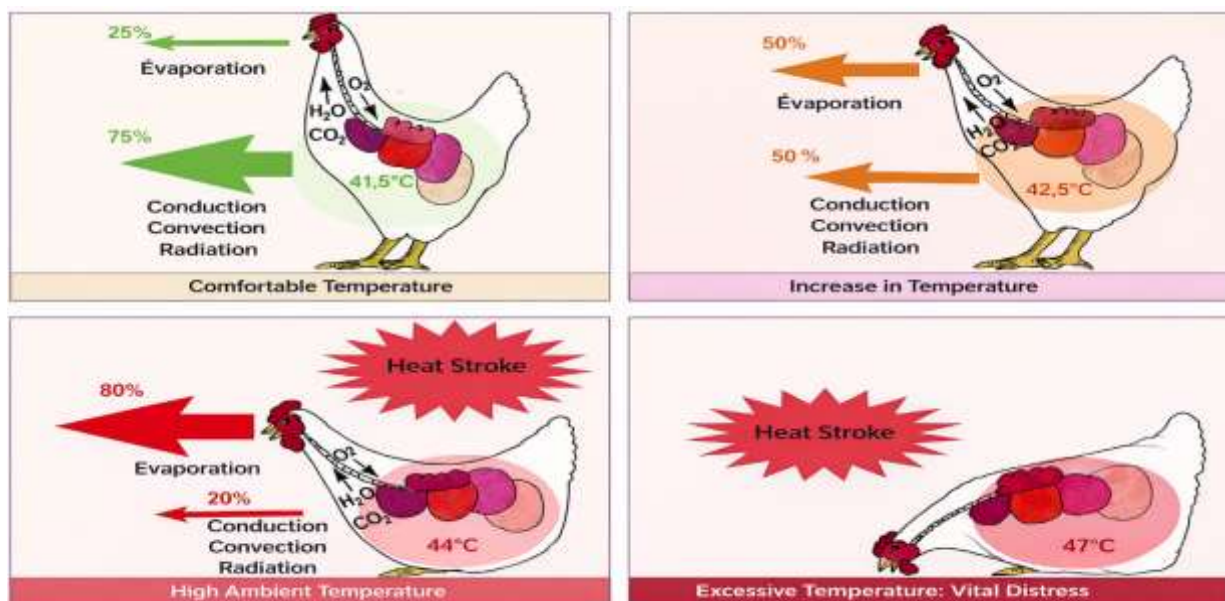


Figure 14: The four phases of heat thermal stress

6. PREVENTION

6.1. FEED

Feeding is an essential factor in heat stress prevention.

- Providing animals with good quality feed means:
- Limiting protein use and optimizing amino acids particularly lysine and methionine;
- More lipids (developing less extra heat during digestion) and fewer carbohydrates, which reduces heat production;
- Using feed additives to improve cellular defenses and minimize damage at the intestinal level;
- Using antipyretics to reduce body temperature;
- Increasing availability of antioxidants through feeding, such as vitamin C and vitamin E.

Example

Adapting feeding times is a good way to provide support to animals in hot climate.

- Give animals 1/3 of the daily ration between 6 a.m. and 10 a.m.
- Then remove feed during the hottest hours of the day (from 10 a.m. to 4 p.m.).
- Give animals the remaining 2/3 of the daily ration between 4 p.m. and 9 p.m.

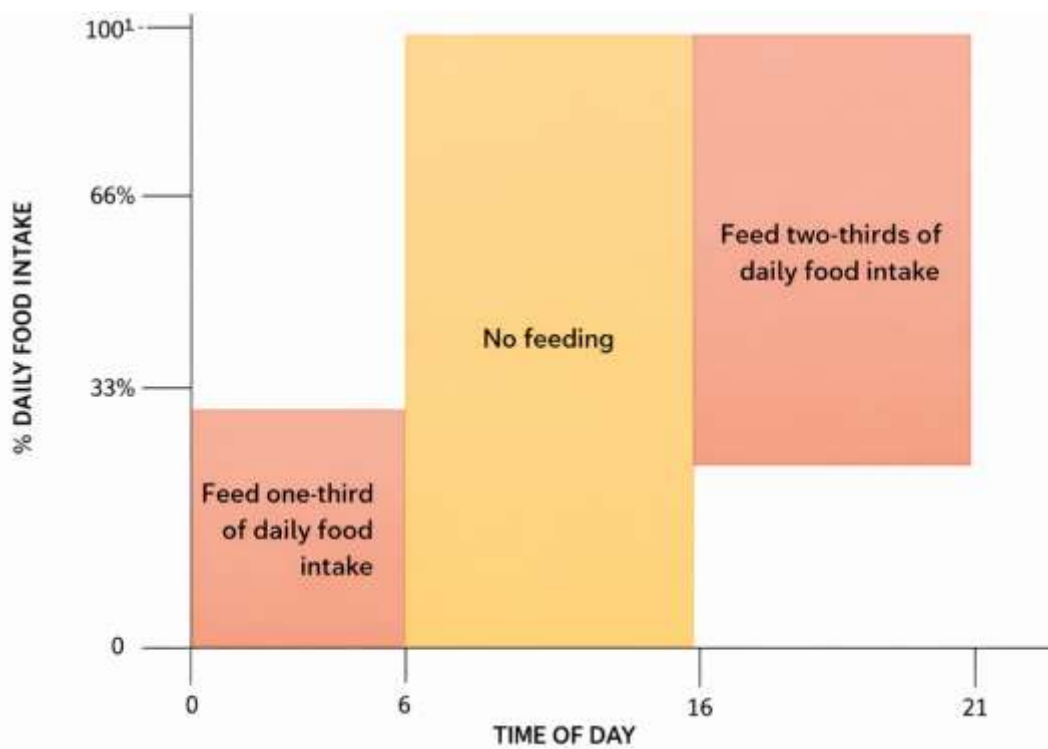


Figure 15: Feed distribution times during heat thermal stress

6.2. WATERING

- Ensure that drinking water is available in abundance and is fresh,
- Easy access to clean water, without germs, and at a temperature lower than core body temperature is necessary,
- Sufficient number of waterers must be provided,
- Use oral anti-stress (vitamin C).

6.3. MANAGEMENT AND CLIMATE

Other good management measures are:

- To prevent heat from entering, building insulation must concern the roof and walls, not the floor.
- To evacuate heat from the building, the installation of tunnel ventilation with an air speed of 2 m/sec is recommended.
- When humidity rate is high, it is not recommended to use water evaporative cooling devices (for example a humidifier).

7. WORK PROTOCOL

7.1. WORKPLACE: at the farm level / farm hens or turkeys.

7.2. NECESSARY EQUIPMENT: surgical alcohol, cotton, electronic thermometer, stethoscope, syringe, glucometer.

7.3. WORK IMPLEMENTATION: expose hens to acute stress (loud noise, hen restraint).

7.4. ASSESSMENT OF THE ANIMAL'S RESPONSE TO STRESS:

Identify behavioral changes in hens, according to the animal's perception of the situation.

Measure physiological changes by:

7.4.1. Non-invasive method

A. Necessary equipment: electronic thermometer and stethoscope.

B. Temperature measurement: in poultry, temperature is taken at the cloaca level by introducing an electronic thermometer into the orifice, so as to touch the internal mucosa.

Physiological norms for temperature in an adult hen are between 40.5 and 42.5 degrees Celsius, with an average of 41.5 degrees Celsius.

C. Heart rate measurement: in hens, a stethoscope can be used to count the number of heartbeats. Heart auscultation is done ventrally.

Heart rate can be measured for 15-30 seconds. Physiological norms for heart rate in hens are between 180 and 340 beats per minute.

D. Respiratory rate measurement: in hens, respiratory rate is measured by counting the number of inspiration and expiration movements observed at the ventral level, over a duration of one minute.

Physiological norms for respiratory rate in adult hens are between 15 and 30 movements per minute. For pulmonary auscultation, a stethoscope can be used at the dorsal level.

It is important to compare the obtained results with the physiological norms of poultry under normal conditions.

7.4.2. Invasive method

A. Necessary equipment: alcohol, cotton, insulin syringe (small format), glucometer.

B. Blood sampling: blood sampling in hens is performed at the wing vein (brachial vein) or saphenous vein level.

The wing vein is located on the inner side of the wing, between the humerus and radius joint. It is advisable to slightly clear the area for better visibility (remove small feathers).

Before sampling, it is necessary to disinfect the area using alcohol, then, the syringe needle is positioned at a 45° angle and carefully introduced into the vein. It is important to aspirate blood gently in order not to compromise the vein.

Blood glucose in a fed hen is between 2 and 3 g/l.



Figures 16 : Temperature measurement at the cloaca level in hens (personal photographs)



Figures 17 : Blood sampling at the wing vein level in hens (personal photographs)

PRACTICAL WORK 3: SLAUGHTER STRESS TEST IN CATTLE

SPECIFIC OBJECTIVES

At the end of this chapter, you will be able to:

- Define standards related to animal welfare during the pre-slaughter period.
- List the slaughter stages in a slaughterhouse.
- Identify pre-slaughter stress factors in cattle.
- Determine cattle responses to pre-slaughter stress.
- Assess the consequences of stress factors on meat quality.

1. GENERAL INFORMATION ON SLAUGHTER

1.1. DEFINITION OF SLAUGHTERHOUSE

It is a specialized establishment, approved by official veterinary services, where animals intended for human consumption are slaughtered and inspected.

1.2. DEFINITION OF SLAUGHTER

Slaughter generally refers to the killing of farm animals intended for the production of meat, fur, wool, and hides.

1.3. DEFINITION OF MEAT

All parts of bovine, ovine, caprine, camelid, and equine species animals, recognized as fit for human consumption with different forms of presentation namely cut meat, deboned or not.

- Meat is one of the richest foods in animal proteins, which results from muscle transformation after slaughter.
- Meat transformation goes through three stages (panting, rigor mortis, and maturation).
- The transformation of muscle into meat involves biochemical mechanisms that affect muscle composition and structure, and consequently especially influences muscle acidity which, in turn, conditions the organoleptic and technological quality of meat.

2. PRE-SLAUGHTER PERIOD

It begins with animal preparation on the farm and ends at the moment of the animal's death after bleeding.

During this period, animals are subjected to a certain number of treatments related to essential operations (change of physical and social environment) which are:

- Fasting (water diet),
- Loading animals into the means of transport,
- Transport (depending on the distance between farm/livestock market and slaughterhouse),
- Unloading animals from the means of transport to the rest area in the slaughterhouse,
- Bringing to the slaughter room.

2.1. TRANSPORT OF ANIMALS TO THE SLAUGHTERHOUSE

Transport is an unavoidable stage in the animal's life when it comes from another farm or market, or when being conveyed to the slaughterhouse.

- Vehicle layout and equipment must be adapted according to the transported species.
- When transporting animals intended for slaughter, it is important to protect their health status and their welfare as much as possible.
- It is imperative to ensure animal comfort during transport in order to minimize risks of injuries and stress.
- At the time of loading animals, the transporter is responsible for guaranteeing their good condition, their correct identification, as well as possession of required health documents.
- Transporters must hold a movement certificate issued by the official veterinarian and follow training for animal handling and transport.
- Safety of animal unloading at the slaughter establishment level is facilitated by the presence of non-slip floors, moderate slope, and side walls (landing area).
- After each transport, it is mandatory to wash and disinfect trucks.

3. STAGES OF ANIMAL SLAUGHTER AT THE SLAUGHTERHOUSE

Good animal handling by operators contributes to reducing animal stress and suffering and ensuring human safety.

3.1. UNLOADING

- At the slaughterhouse, animals are unloaded from the transport truck calmly, with adapted docks (low slope) with anti-slip flooring.
- While ensuring their own safety, slaughterhouse operators must avoid any stress, injuries, or pain to animals.
- Animals should not be forced to move faster than their normal pace in order to avoid injuries following falls or slips.

3.2. RECEPTION / CONTROL

- Upon animal reception, the person in charge controls traceability through identification tags, which must correspond to their movement document.

3.3. ANIMAL CARE AND ANTE-MORTEM INSPECTION

- Animals are then placed in the rest area, which is equipped with waterers and arranged to facilitate their movement and then their rest.
- The floor of the unloading platform, corridors, and holding areas must be made of non-slip materials and well maintained to minimize animal slips as much as possible.

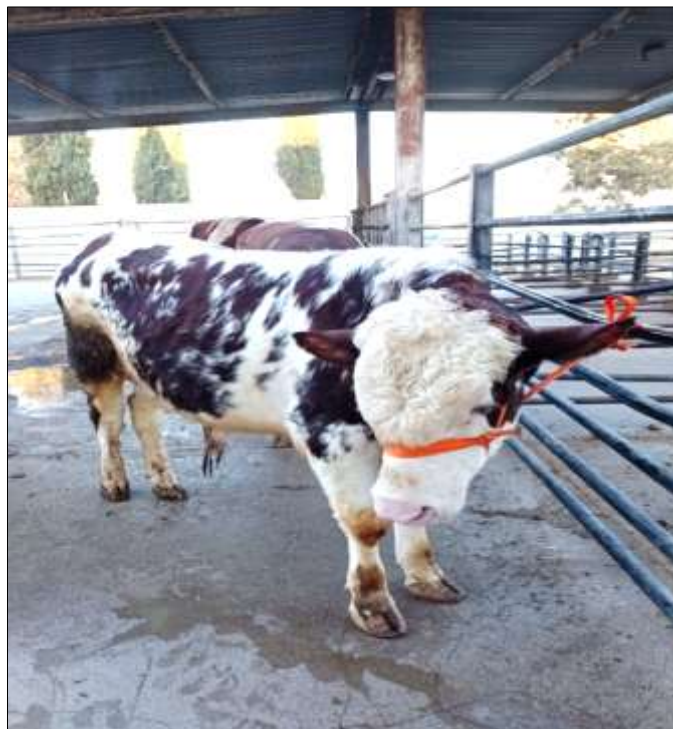


Figure 18: Placing in holding pens (rest area), (personal photograph)

- Veterinary inspectors verify the health status of animals: this is the ante-mortem inspection.
- Animals in poor condition (signs of disease, fatigue, lameness, injury...) are isolated in the detention pen.

3.4. DRIVING

- When driving groups of animals to the slaughter station, everything is implemented to avoid their stress, notably with the presence of non-slip floors or anti-backup devices to avoid jostling.

3.5. RESTRAINT

- Restraint is mandatory to facilitate animal slaughter and to protect the handler.
- Animal restraint is performed using adapted equipment to allow their immobilization.

Note

In non-Muslims before the bleeding stage, stunning is performed using a slaughter gun, which triggers the animal's immediate loss of consciousness.



Figure 19: Cattle restraint before bleeding (personal photograph)

3.6. BLEEDING

- In Muslims, slaughter is performed without prior stunning, which makes animal restraint (head and body) mandatory.
- Animal immobilization must be rapid, of short duration, and throat cutting must be performed immediately without delay.

- The operator therefore directly performs bleeding, which consists of severing the main blood vessels and tissues of the cervical region.
- Blood loss following bleeding will induce loss of consciousness then death of the animal.

3.7. SKINNING

- Once the animal is dead, the hide is separated from the carcass, manually or semi-automatically by a machine that remains guided by a knife.
- This is followed by removal of the head and limbs.

3.8. EVISCERATION

- This stage consists of removing all thoracic and abdominal viscera from the animal.

3.9. SPLITTING

- Bovine carcasses are then split in two for commercial constraints and to facilitate sanitary inspection.

3.10. POST-MORTEM INSPECTION

- Official veterinary services then control the sanitary conformity of the carcass and offal and seize those that are unfit for consumption, then they order stamping for their marketing.

3.11. WEIGHING / GRADING

- Carcasses are prepared to be weighed and graded. They can thus be evaluated according to their weight.

3.12. CARCASS REFRIGERATION

- Carcasses and offal are stored in a cold room, where carcasses rest for at least 24 hours to reach optimal cooling and mature for a variable duration.
- This involves applying temperatures as low as possible, while remaining above freezing point, in order to guarantee adequate preservation.

Note: According to different marketing channels, meat can be delivered directly in carcass form, quarters, or prepared and packaged in trays, particularly if the slaughterhouse has a meat product processing unit.

4. STRESS AND STRESS FACTORS

4.1. SLAUGHTER STRESS

Slaughter comprises a series of potentially stressful procedures (pre-slaughter period); these generally begin with animal fasting, transport to the slaughterhouse, and end with the animal's killing.

- The conditions to which animals are subjected during this pre-slaughter period are all sources of stress that can significantly affect meat quality.

4.2. STRESS FACTORS

4.2.1. Stress factors of physical or physiological origin

- Food deprivation (water diet),
- Fatigue (animal transport),
- Overcrowding for a long duration,
- Poor handling,
- Slips, falls, and injuries.

4.2.2. Stress factors of psychological origin

- Human presence,
- Pain,
- Fear,
- Absence of familiar conspecifics.

4.2.3. Stress factors of behavioral origin

- Lack of exercise, prolonged standing position.

4.2.4. Stress factors of environmental origin

- Confrontation with new events,
- Environment change,
- Ambient temperature variation, noise and congestion,
- Type of transport vehicle handling (acceleration, braking...).

5. MAIN STRESS RESPONSES

5.1. PHYSIOLOGICAL RESPONSES

- Correspond to increased heart and respiratory rate, and stress hormone secretion.
- Increased frequency of urination and/or defecation.
- Increased blood glucose concentration, blood pressure, and muscle tone.
- Increased blood concentration of cortisol and ACTH, and glucagon are frequently used to assess animal stress level.

5.2. BEHAVIORAL REACTIONS

- Can manifest as attacks (aggressiveness).
- Escape attempts.
- These changes are used to assess animal stress level. Moreover, they facilitate interpretation of physiological measurements.

Note

These physiological and behavioral reactions can have measurable effects on meat quality, whose mechanisms involve muscle energy metabolism.

Post-mortem muscle metabolism, as well as temperature and pH evolution, allow appreciation of the level of physical activity, and sometimes stress of the animal before its slaughter.

6. CONSEQUENCES OF STRESS FACTORS

Can vary according to these characteristics:

- Nature,
- Intensity,
- Duration of stimulus,
- But also according to the nature of the animal's perception. Perception which will evolve according to age, sex, and genetics.

6.1. BEHAVIORAL AND PHYSIOLOGICAL REACTIONS OCCURRING IN THE MINUTES PRECEDING SLAUGHTER (ACUTE STRESS)

Stress and physical activity at the moment of the animal's killing accelerate glycogen depletion.

Resulting in an acceleration of muscle metabolism that persists after death and which can manifest as: faster post-mortem muscle acidification (pH drop), while muscle temperature decrease is slowed.

pH can be below 6 within 45 minutes after slaughter.

It is the initial combination of low pH and high temperature that leads to protein denaturation.

These proteins become unable to retain muscle water, which gives meat with low water holding capacity (wet cut) and becomes tough after cooking.

The meat will then be paler, with poorer water holding capacity and is called pale, soft, exudative (PSE) meat. It is feverish meat.

Note

In cattle, the higher the heart rate during the minutes preceding slaughter, the faster the muscle pH decrease.

6.2. BEHAVIORAL AND PHYSIOLOGICAL REACTIONS OCCURRING IN THE HOURS PRECEDING SLAUGHTER (CHRONIC STRESS)

Animal stress associated with physical activity (fatigue before slaughter) leads to a decrease in muscle glycogen reserves, which causes low lactic acid production, which can result in higher pH and darker meat color.

Muscle proteins are not denatured, since they retain their water holding capacity (dry cut), the meat is characterized by firm texture and lower tenderness.

The meat becomes sticky. It is called dark, firm, dry (DFD) meat.

It is overworked meat, which is not appreciated by consumers and whose shelf life is shorter. This induces meat depreciation with economic consequences.

Note

In cattle, it is well established that certain slaughter conditions, such as mixing animals (livestock market) or long-duration transport increase the risk of producing high-pH meat.

7. PREVENTION

- In order to avoid pH increase, it is necessary to reduce:
- Fighting (physical expenditure),
- Mixing of animal batches, brutal unloading (stress),
- Large temperature differences.

For this, it is necessary to:

- Unload animals calmly, quickly upon truck arrival (reduce waiting),
- Not mix males and females in pens as well as animals with or without horns,
- Plan rapid slaughter if transport has been constraining.

8. WORK PROTOCOL

8.1. WORKPLACE: at the farm/cattle farm level

8.2. NECESSARY EQUIPMENT: surgical alcohol, cotton, electronic thermometer, stethoscope, syringe, glucometer.

8.3. WORK IMPLEMENTATION: expose cattle to acute stress (loud noise, presence of strangers).

8.4. ASSESS THE ANIMAL'S RESPONSE TO STRESS

- Identify behavioral changes in cattle such as flight, aggressiveness, fear according to the animal's perception of the situation.

8.4.1. Non-invasive method:

A. Necessary equipment: thermometer and stethoscope.

B. Temperature measurement: in cattle, temperature is taken rectally by introducing an electronic thermometer into the anal orifice so as to touch the rectal mucosa.

- Physiological norms for temperature in adult cattle are between 38 and 39 degrees Celsius, while in young cattle, they are between 38.5 and 39.5 degrees Celsius.

C. Heart rate measurement: in cattle, a stethoscope can be used to count the number of heartbeats.

- Simply place the stethoscope under the tip of the elbow, that is, between the humerus and the rib cage. Heart rate can be measured over a period of 30 to 60 seconds.
- Physiological norms for heart rate in adult cattle are between 60 and 90 beats per minute, while in young cattle, they vary between 90 and 110 beats per minute.

D. Respiratory rate measurement: in cattle, respiratory rate measurement is done by counting the number of inspiration and expiration movements observed at the flank hollow level, particularly on the right side, over a duration of one minute.

- Physiological norms for respiratory rate in cattle are between 15 and 35 movements per minute.
- Compare the obtained results with physiological norms specific to cattle under normal conditions.

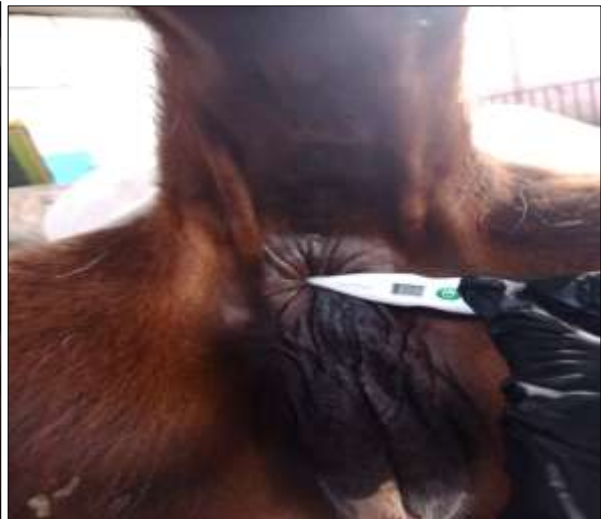
8.4.2. Invasive method:

A. Necessary equipment: alcohol, cotton, syringe, glucometer.

B. Blood sampling: blood sampling in cattle is generally performed from the jugular vein, located at the animal's neck level, or from the caudal vein located at the tail level.

C. Sampling method: to collect blood from the jugular vein, pressure is applied downstream in order to locate the vein.

- Before sampling, it is essential to disinfect the area using alcohol.
- Then, the syringe needle is positioned at a 45° angle and delicately introduced into the vein, followed by gentle blood aspiration.
- Blood glucose in cattle is between 0.4 and 0.7 g/l.



Figures 20 : Rectal temperature measurement in cows (personal photographs)



Figure 21 : Heart rate measurement in cows (personal photograph)

Figure 22 : Respiratory rate measurement in cows at the flank hollow level (personal photograph)



Figure 23 : Localization of the jugular vein in cows (personal photograph)

Figure 24 : Localization of the caudal vein in cows (personal photograph)

PRACTICAL WORK 04: STRESS TEST AND RESPONSE ASSESSMENT IN HORSES

SPECIFIC OBJECTIVES

At the end of this chapter, you will be able to:

- Know the behavioral characteristics of horses.
- Identify types of stress in horses.
- Differentiate between stress factors in horses.
- Verify the horse's response to stress.
- Assess the horse's response to stress.

1. BEHAVIORAL CHARACTERISTICS OF HORSES

- Horses are gregarious animals that function with a group hierarchy. Whatever their place in the herd, horses are reassured by their relationships with other group members.
- The horse is a naturally anxious (stressed) animal, because of its prey status in the wild, which multiplies potential stress sources.
- Before its domestication, its survival depended on its ability to flee in time and as quickly as possible.
- It is therefore an animal whose instinct keeps the senses alert, which preferably chooses to graze in a vast and open place in order to be able to monitor potential predators.
- Domestic horses live very differently from their ancestors. They often live in stables, where their social interactions and movements are restricted throughout the day.

2. STRESS AND TYPE OF STRESS

Stress is at the origin of a reaction that allows all living beings, including horses, to react to difficult situations. It is therefore completely normal, natural, and even useful.

Stress becomes a danger when it is too great.

When talking about stress, a distinction is often made between acute stress and long-term chronic stress (related to the notion of time).

2.1. ACUTE STRESS

In case of acute stress, the horse's reaction is rapid and of short duration; the latter recovers quickly, because the stress disappears with the disappearance of the stress factor.

Example

- If the horse is frightened by a plastic bag, sudden noise, it will startle or possibly shy and then resume its activities.

2.2. CHRONIC STRESS

In case of chronic stress, this recovery does not occur or will occur after a long period.

Example

- During a move to a new stable (environment change), the horse may be stressed for a long period, the time to adapt to all the new things.
- It can also experience so many moments of acute stress (repeated stress) that its organism cannot recover sufficiently between the two.

3. STRESS FACTORS

Table 2: Stress-generating factors in horses

Type of stress	Stress factors
Acute stress	Poor management of feed intake Surgery or injury Traumatic transport Over-exhaustion Sudden noise
Chronic stress	Environment change Permanent confinement Boredom Lack of exercise Chronic obstruction of small airways Chronic lameness Back pain Dental problem Prolonged fatigue

4. ANIMAL'S RESPONSE TO STRESS

- ✓ The horse is frightened or placed in a difficult situation for it.
- ✓ The amygdala, the brain's fear center, sends signals via the hypothalamus and pituitary gland to the adrenal glands to produce catecholamines, hormones such as adrenaline and noradrenaline (see Figure 3).
- ✓ These substances prepare the horse's body for action.
- ✓ Which results in behavioral and physiological changes.

4.1. BEHAVIORAL CHANGES

- The horse must choose between three situations: flee, fight, or endure.
- The degree of reaction to a stressful event varies according to the individual (according to the animal's perception of the situation).

4.2. PHYSIOLOGICAL CHANGES

- Heart rate accelerates,
- Blood pressure increases,
- Breathing becomes more shallow,
- More nutrients are brought to muscles, blood glucose supply increases.
- A few minutes after the release of catecholamines in the organism, the hypothalamus, pituitary gland, and adrenal glands will release glucocorticoids: cortisol.
- Faced with a real threat, this endocrine reaction is beneficial, increasing the horse's alertness and energy level so that it can react appropriately.

Note

- ✓ When cortisol level remains elevated over a long period without ever returning to normal, this is a sign of chronic stress.
- ✓ This state can have negative repercussions on the horse's health and behavior.
- ✓ Horses that have a high concentration of this hormone in their blood for long periods may present:
 - Aggressive behavior,
 - Increased risks of cardiovascular problems,
 - Deterioration of immune function,

- Increased risk of gastric ulcers and digestive disorders,
- Reduced growth rate and inhibited reproductive function.

5. ASSESSMENT OF THE HORSE'S RESPONSE TO STRESS

✓ Assessment of stress response in horses is crucial to identify triggering factors and implement mitigation strategies.

- Owners and equine professionals can carefully observe horse behavior, monitor physiological signs such as heart rate and breathing, and assess changes in appetite and body weight.
- Tools such as behavior rating scales and physiological measurements can also be used for more in-depth assessment.

5.1. BEHAVIORAL CHANGES

✓ The stressed horse acts in an unusual and often aggressive manner:

- Ears pinned back,
- Head raised,
- Explosive reactions,
- Dilated nostrils,
- Agitation,
- Excessive sweating,
- Trembling,
- Pawing the ground with hooves,
- Excessive vocalizations,
- Loss of appetite,
- Repetitive movements.

5.1.1. Ear positioning

- Ears pointed backward are a sign of aggressiveness and negative emotion.



Figure 25: The different ear positions in horses according to their emotional state



Figure 26: The different expressions of nostrils and nose tip in horses according to their emotional state



Figure 27: Horse's response to an unusual noise

In Figure 27, the horse is tied.

- It hears an unusual noise coming from outside.
- It feels stress and this is clearly visible in its expression.
- Its eyes are wide open, its nostrils dilated, its nose has a concave shape.
- It points its ears in the direction of what worries it.
- It produces stress droppings.

5.1.3. Development of stereotypies

- Stereotypies develop in response to different stress factors. Among these manifestations, we frequently observe stereotypies such as those related to cribbing, wind-sucking, or weaving in horses.
- These behaviors are particularly common in anxious equids that have difficulty tolerating loneliness, boredom, or confinement.

A. Cribbing:

Is when the horse takes support with its incisors on any support (door, or other), contracts its neck, and emits a noise.

It is a difficult stress to correct. It can be related to gastric disorders. It is often identified by the premature wear of teeth that it causes.



Figure 28: Cribbing in horses

B. Wind-sucking:

The horse swallows air while most of the time taking support on a surface: manger, stall door, post.

Apart from material deterioration, this stereotypy is dangerous for the horse's health.

Indeed, aerophagia is directly correlated with the risk of presenting colic and the presence of gastric ulcers.

Moreover, this causes premature wear of incisors.

C. Weaving:

It consists of swaying from one foreleg to the other from left to right and vice versa. The horse has its head low, nose below knee level.

It swings its head from right to left and its weight shifts from one foreleg to the other. It has vacant eyes, ears are relaxed.

It is less harmful than wind-sucking for the horse's digestive system, but it has harmful effects on the horse's locomotor system, by abnormally stressing the joints and collateral ligaments of the forelegs.

Note

In fact, the horse does this to relax. The stereotypy produces soothing endorphins.

5.1.4. Excessive sweating

- Greater perspiration is also considered a sign of stress.
- In a stress situation, the horse will have the instinctive reflex to flee.

- When it cannot do so or when placed in constraining situations (competitions, transport, stall confinement), its stress will manifest through body reactions: muscle contractions, ear movements, etc.

5.2. PHYSIOLOGICAL CHANGES

Physiological signs are:

- Increased heart rate,
- Increased respiratory rate,
- Increased tension, presence of stress hormones in blood, and increased blood glucose (energy needed to cope with the situation when exposed to acute stress).

5.3. PATHOLOGICAL DISORDERS

If stress lasts a long time, we can observe the development of pathologies.

5.3.1. Gastric ulcers

- Stressed horses are more exposed to developing gastric ulcers.
- Stomach acidity attacks the mucous membranes, which can cause inflammation or even deep lesions (ulcers). This is painful and can cause weight loss, decreased fitness, or colic.
- In case of intense stress, the horse can also present severe diarrhea, which can lead to severe dehydration.

5.3.2. Injuries

- The second danger of stress in horses is the risk of injury. Indeed, the panicking horse can have completely unpredictable and very dangerous reactions for itself.
- Unreasonable fear can lead to a flight instinct, the horse then being capable of throwing itself in front of a car, into a ditch or barrier, or jumping an impassable obstacle.
- We thus observe a greater number of injuries in stressed horses, most affecting the limbs or head, which can lead to very serious injuries.

6. WORK PROTOCOL

6.1. WORKPLACE: at the farm/equine farm level.

6.2. NECESSARY EQUIPMENT: surgical alcohol, cotton, electronic thermometer, stethoscope, syringe, glucometer.

6.3. WORK IMPLEMENTATION: expose a horse to acute stress (loud noise, movement with a bag).

6.4. ASSESSMENT OF THE ANIMAL'S RESPONSE TO STRESS

✓ Identify behavioral changes in the horse at the moment of stress (according to the animal's perception of the situation).

✓ Measure physiological changes by:

6.4.1. Non-invasive method:

A. Necessary equipment: electronic thermometer and stethoscope.

B. Temperature measurement: in horses, temperature is taken rectally by introducing an electronic thermometer into the anal orifice, so as to touch the rectal mucosa.

✓ Physiological norms for temperature in an adult horse are 36.5 to 38 degrees Celsius.

C. Heart rate measurement: in horses, a stethoscope can be used to count the number of heartbeats.

✓ Simply place the stethoscope under the tip of the elbow, that is, between the humerus and the rib cage. Heart rate can be measured over a period of 30 to 60 seconds.

✓ Physiological norms for heart rate in an adult horse are between 28 and 44 beats per minute, while in young horses, they vary between 50 and 70 beats per minute.

D. Respiratory rate measurement: in horses, respiratory rate measurement is done by counting the number of inspiration and expiration movements observed at the abdominal level, and recording them for one minute.

✓ Physiological norms for respiratory rate in adult horses are between 8 and 16 movements per minute, while in young horses, they vary between 10 and 20 movements per minute.

✓ Compare the found results to the horse's physiological norms under normal conditions.

6.4.2. Invasive method:

A. Necessary equipment: alcohol, cotton, syringe, glucometer.

B. Blood sampling: blood sampling in horses is generally performed from the jugular vein, located at the jugular groove level between the brachiocephalic muscle and the sternocephalic muscle.

C. Sampling technique: pressure is applied downstream to locate the vein.

✓ The sampling area is disinfected using alcohol, following the direction opposite to hair growth.

✓ Then, the syringe needle is positioned at a 45° angle and delicately introduced, followed by blood aspiration.

✓ Blood glucose in horses is between 0.74 and 1.15 g/l.



Figure 29: Rectal temperature measurement in horses (personal photograph)



Figure 30: Heart rate measurement in horses (personal photograph)



Figure 31: Locating the jugular vein (personal photograph)

