



University of Novi Sad
Faculty of Agriculture



ANTIMICROBIAL ACTIVITY OF GARLIC (*A. SATIVUM* L.) AGAINST *SALMONELLA ENTERITIDIS* AND *STAPHYLOCOCCUS AUREUS*

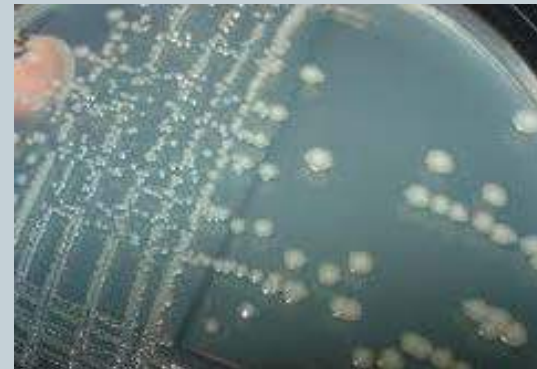
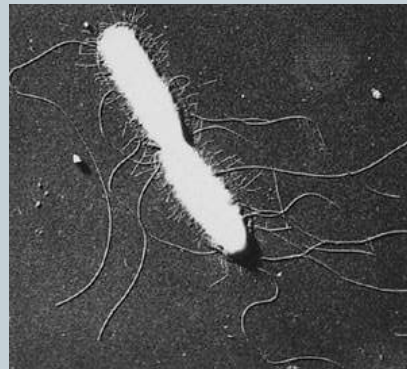
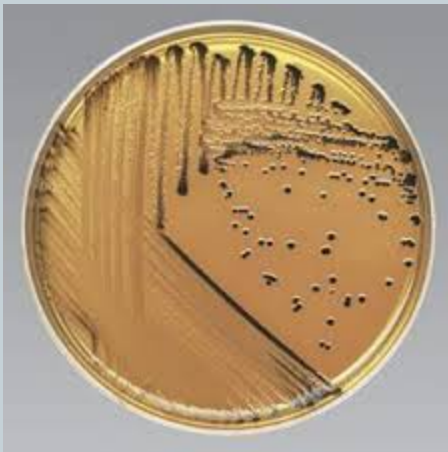
*SLOBODAN VLAJIĆ, LJILJANA KURUCA,
STANISLAV SIMIN, ZORA JELESIĆ,
MIRA MIHAJLOVIĆ-UKROPINA, ANA TAKAČ,
VESNA LALOŠEVIĆ*



INTRODUCTION – *Salmonella Enteritidis*



Salmonella Enteritidis is a serotype of *Salmonella enterica* subsp. *enterica* which is, together with *Salmonella Typhimurium*, most frequently associated with human illness



INTRODUCTION - *Salmonella Enteritidis*



- Common source of infection: contaminated eggs and poultry meat



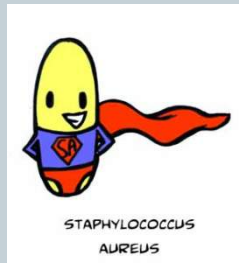
- Disease:

- Gastrointestinal
- Bacteriemia and meningitis



- Asymptomatic carriers!!!

INTRODUCTION – *Staphylococcus aureus*



Gram-positive, ubiquitous cocci

Bacterial meningitis

- *Streptococcus pneumoniae*
- *Neisseria meningitidis*
- *Haemophilus influenzae*
- *Streptococcus agalactiae*
- *Listeria monocytogenes*

Otitis media

- *Streptococcus pneumoniae*

Pneumonia

Community-acquired:

- *Streptococcus pneumoniae*
- *Haemophilus influenzae*
- *Staphylococcus aureus*

Atypical:

- *Mycoplasma pneumoniae*
- *Chlamydia pneumoniae*
- *Legionella pneumophila*

Tuberculosis

- *Mycobacterium tuberculosis*

Skin infections

Eye infections

- *Staphylococcus aureus*
- *Neisseria gonorrhoeae*
- *Chlamydia trachomatis*

Sinusitis

- *Streptococcus pneumoniae*
- *Haemophilus influenzae*

Upper respiratory tract infection

- *Streptococcus pyogenes*
- *Haemophilus influenzae*

Gastritis

- *Helicobacter pylori*

Food poisoning

- *Campylobacter jejuni*
- *Salmonella*
- *Shigella*
- *Clostridium*
- *Staphylococcus aureus*
- *Escherichia coli*

Sexually transmitted diseases

Urinary tract infections

Diverse spectrum of human and animal diseases ranging from minor skin infections to life threatening conditions, such as pneumonia and meningitis



INTRODUCTION – Antimicrobial resistance

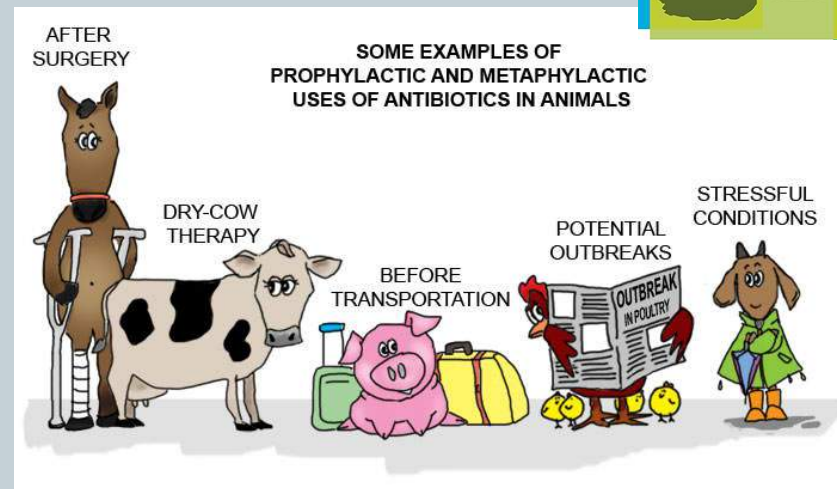
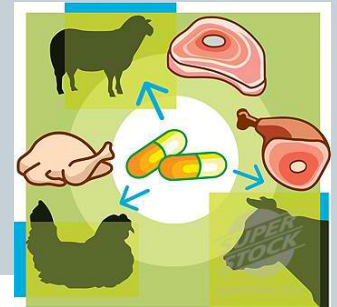


- Worldwide, number of reports on antimicrobial resistance in both human and animal bacteria is increasing

- The resistance develops as a consequence of:

Extensive prophylactic and therapeutic usage of antimicrobial drugs, often in subtherapeutic concentrations

Their administration as growth promoters in food animal production



INTRODUCTION – Antimicrobial resistance



- Consequently, antimicrobial-resistant bacteria in food animals may threaten the efficacy of human drugs



A need for an **alternative** approach!



Results of studies on complementary and alternative medicine practices suggest that introduction of plant extracts, such as garlic extract, in antimicrobial therapy may significantly decrease this emerging burden of drug resistance

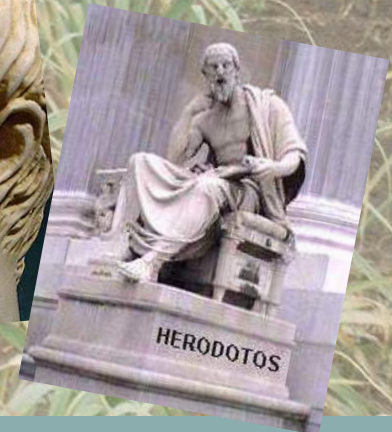
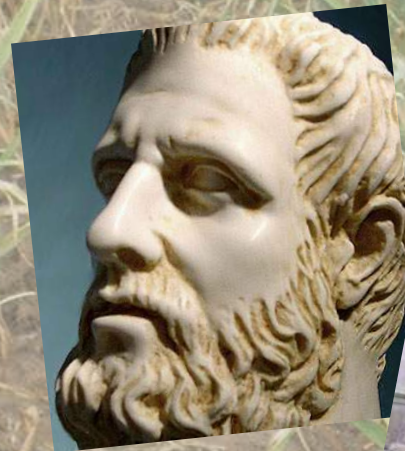
INTRODUCTION – *Allium sativum* L.



Hardy annual monocotyledon plant, and one of the oldest cultivated vegetable crops



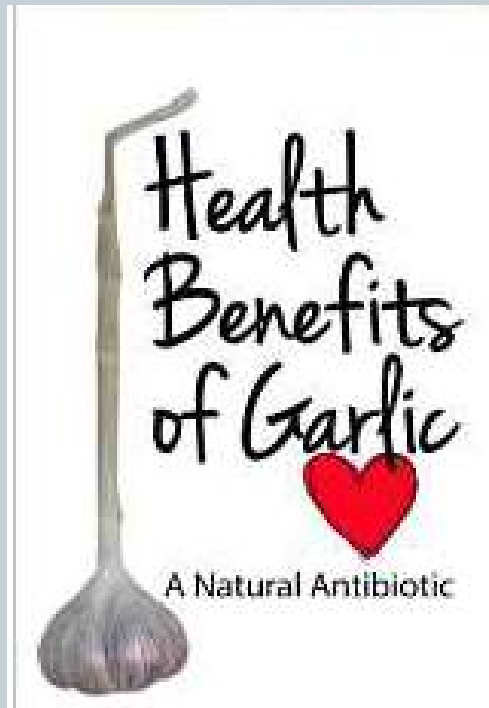
The earliest record of the medical properties of garlic and directions for its use come from historian Herodotus and Hippocrates



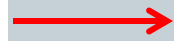
INTRODUCTION – *Allium sativum* L.



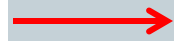
- **Alicin!**



Antibiotic



Antidiabetic



Hepatoprotective



Antioxidant



Anticancer?



MATERIAL AND METHOD OF THE STUDY

- **Bacteria:**

- test sample consisted of 9 human isolates of *S. aureus* and 13 isolates of *S. Enteritidis*
- Bacterial suspensions in saline (6×10^8 cfu/ml, the 2nd scale according to McFarland) were made for the inoculation of Muller-Hinton agar

- **Garlic extract:**

- winter garlic (*Allium sativum* var. *vulgare*) cloves were immersed in 75% alcohol and rinsed with water
- Crude garlic extract was obtained using mechanical compression of the cloves

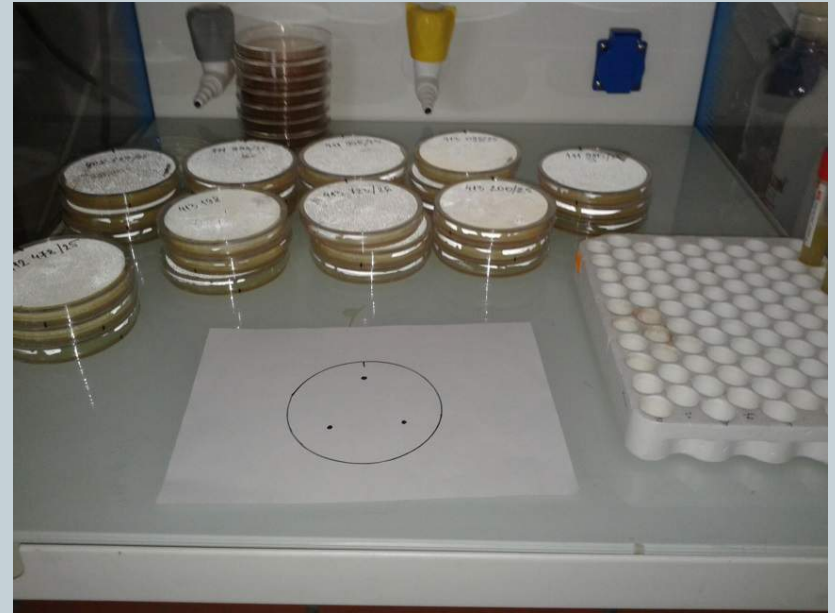


MATERIAL AND METHOD OF THE STUDY



- **Disk-diffusion method:**

- On each inoculated plate, three filter papers (R=6mm) were placed:
 1. Concentrated garlic extract
 2. 50% solution of the extract
 3. Sterile distilled water (control)
- Plates were incubated for 20h on 37°C

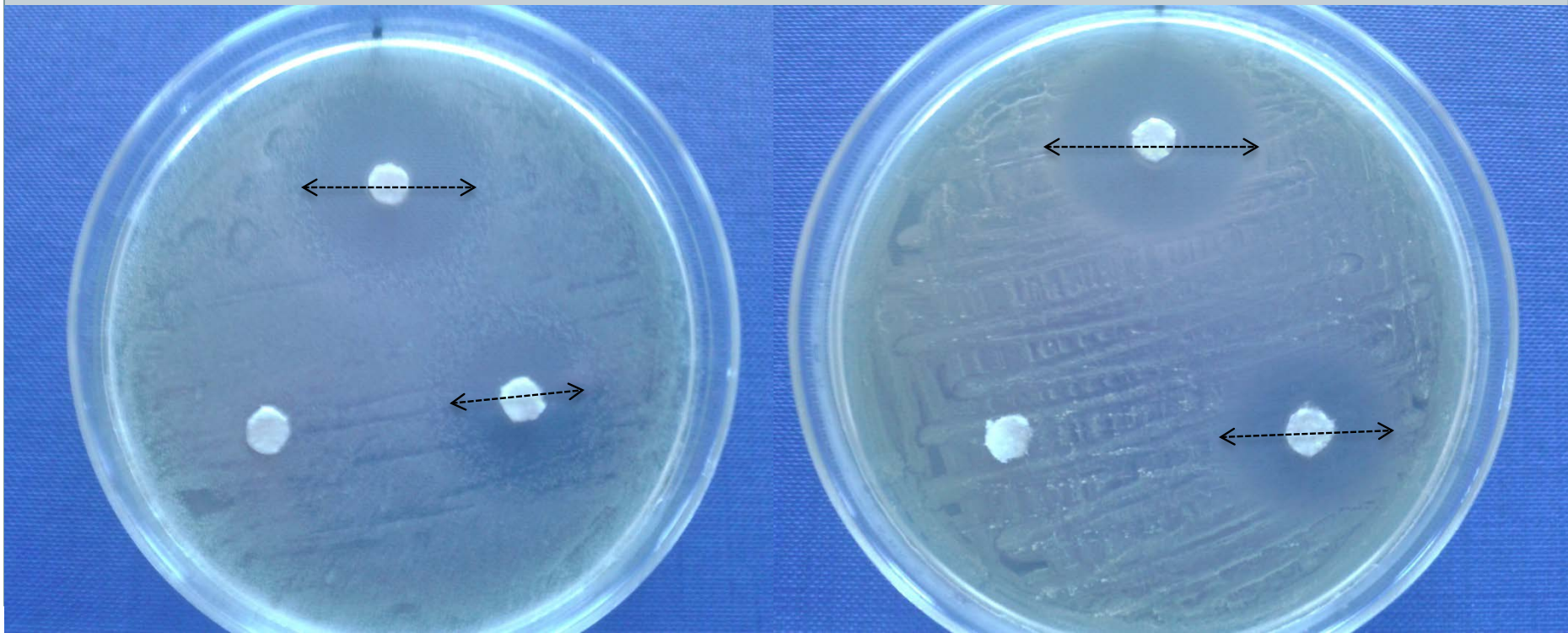


Test was set in 3 replications

Results were analysed in Statistica 10 software

RESULTS and DISCUSSION

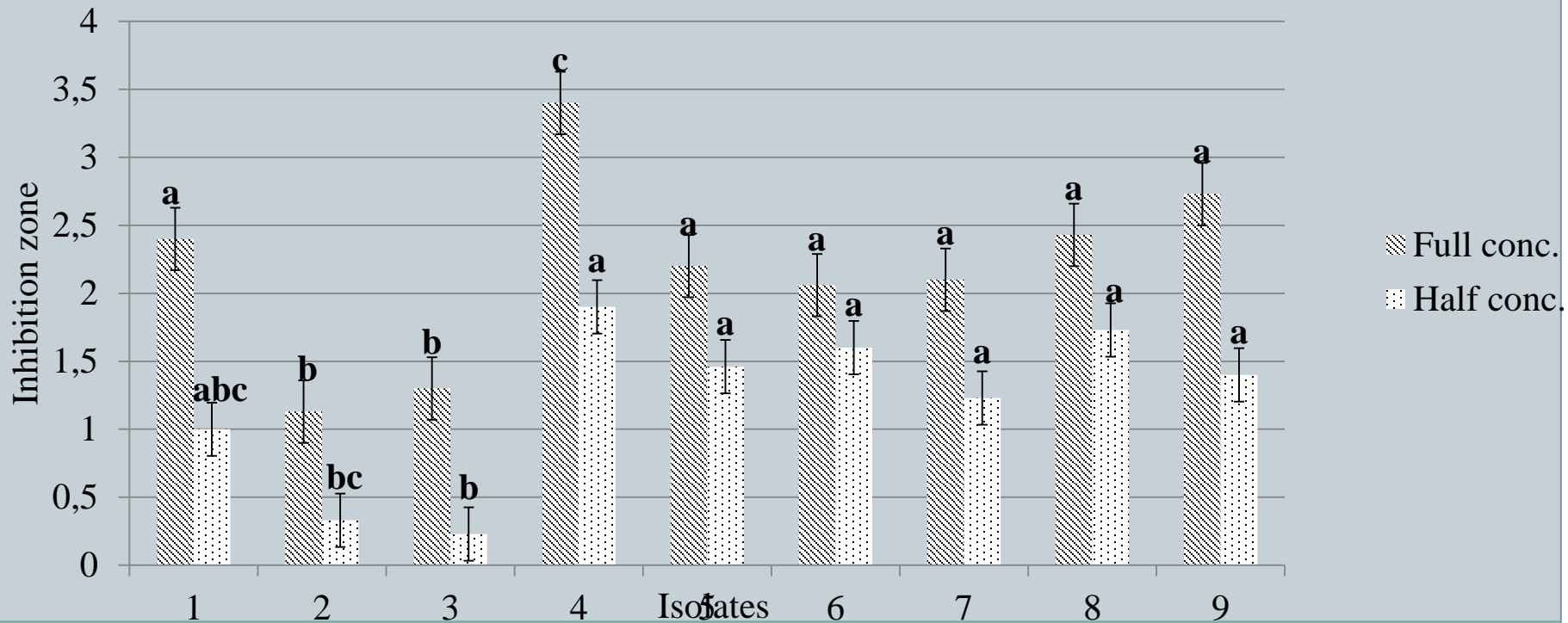
Extract concentration	Average inhibition zones of bacteria (mm)		p-Value
	<i>S. aureus</i>	<i>S. Enteriditis</i>	
100%	21.6	21.4	0.940
50%	12.1	11.6	0.815



RESULTS and DISCUSSION

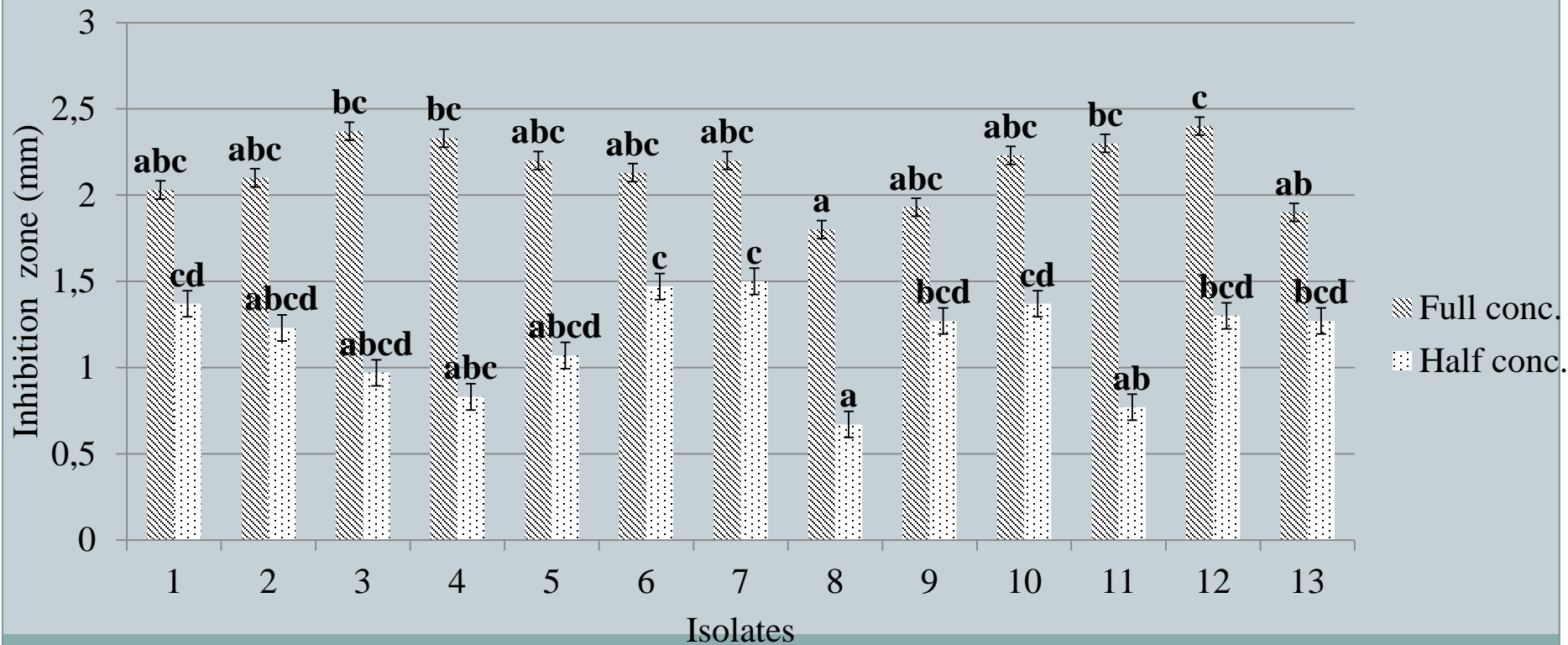


Comparison of susceptibility of isolates within the
S. aureus species



RESULTS and DISCUSSION

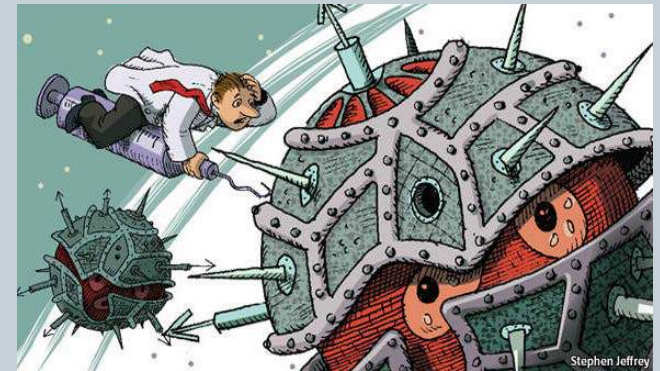
Comparison of susceptibility of isolates within the
S. Enteritidis serotype



RESULTS and DISCUSSION



- According to the European Food Safety Authority (EFSA) and European Centre for Disease Prevention and Control (ECDC), a statistically significant **decrease in the number of human salmonellosis cases** was observed in the EU
- On the other hand, an **increase in antimicrobial resistance** of this pathogen is being reported worldwide
 - ✦ In Serbia, resistant strains of *S. Enteritidis* have been confirmed in both human and animal samples!
- Salmonella serotype with the lowest antimicrobial resistance seems to be *S. Enteritidis*
- Fortunately, resistance to the critically important antimicrobials for human medicine, cefotaxime (a third-generation cephalosporin) and ciprofloxacin (a fluoroquinolone) is , **currently**, relatively low



RESULTS and DISCUSSION

- In staphylococci, the **methicillin-resistance** is considered to be the most significant form of antimicrobial resistance

- Until recently, human isolates of methicillin-resistant *S. aureus* (MRSA) were usually associated with **hospital-acquired infections**; currently, it became evident that MRSA may also represent a challenge as both **community-acquired infections and colonizing zoonotic agent in companion and food producing animals**

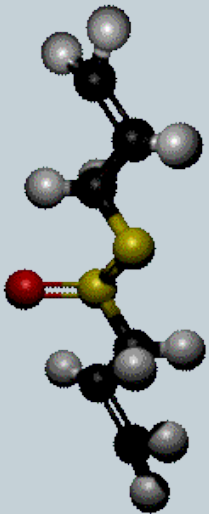


RESULTS and DISCUSSION

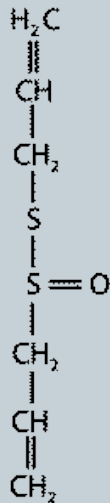
Garlic has been described as **one of the most significant plants of complementary and alternative medicine.**

It holds up the highest concentration of sulfur compounds, such as **allicin**, which are considered **responsible for its anti-microbial activity**

Allicine



Ball-and-stick model of the (R)-allicin molecule



other names
diallylthiosulfinaat;
diallyldisulfide-S-oxide;

Molar mass : 162.27 g mol⁻¹

Density : 1.112 g cm⁻³

Molecular formula
C6H10OS2

The mechanism of antibacterial activity of garlic has not been fully elucidated, however, it is assumed that the primary tool of allicin is the **blocking of RNA formation.**

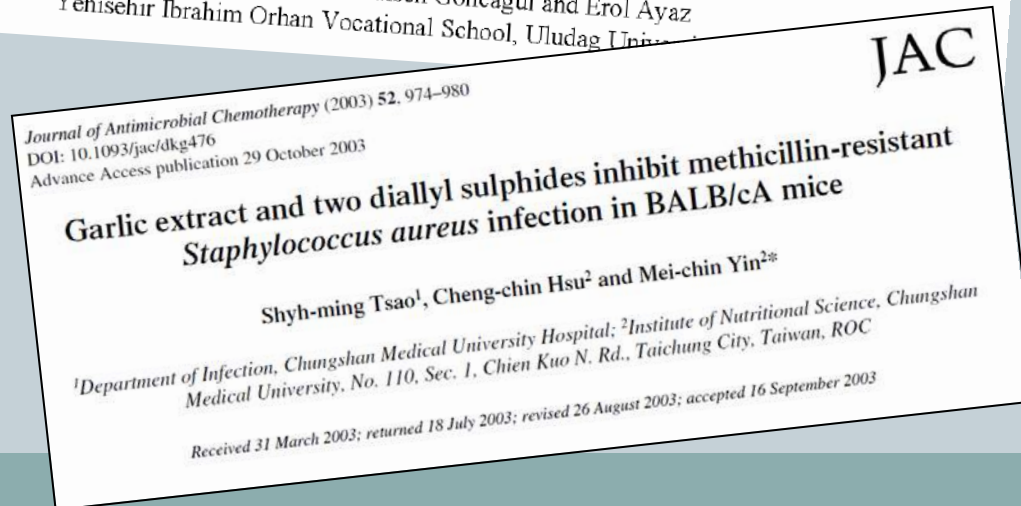
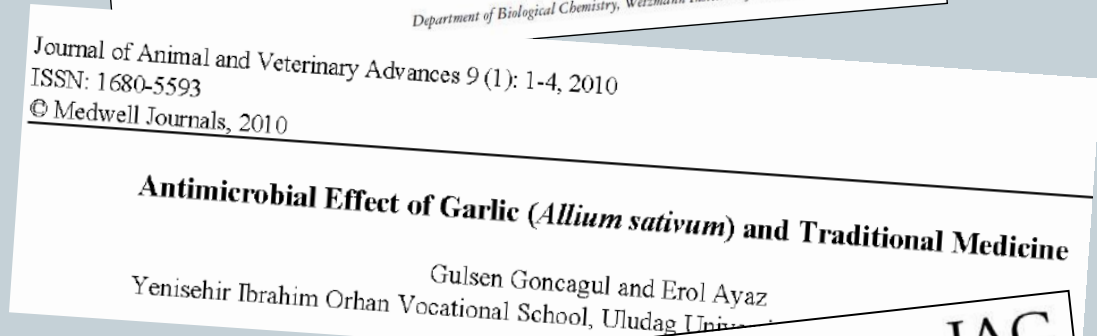
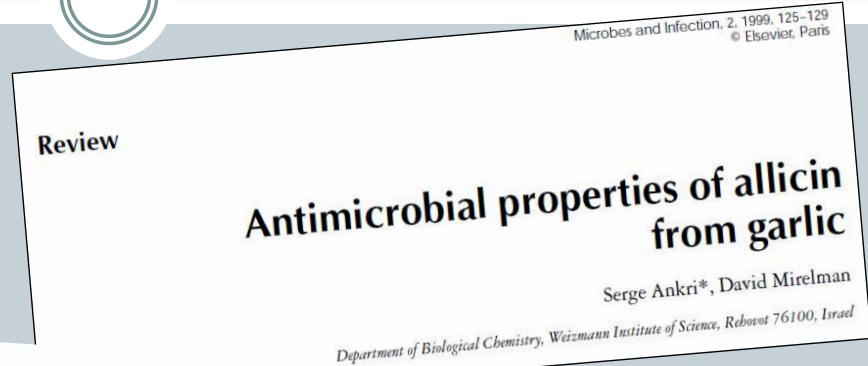
RESULTS and DISCUSSION

- Antimicrobial efficacy of garlic against *Salmonella* and *Staphylococcus*, including the resistant strains, has been observed by authors around the world, in both *in vitro* and *in vivo*

- All these studies reported a concentration/dose-dependent antimicrobial efficacy of garlic compounds, which is in concordance with our results.

- Synergistic action of garlic in combination with some antibiotics was observed

- Development of resistance to beta-lactam antibiotics was 1000-fold easier than development of resistance to allicin from garlic!



CONCLUSION



Both *S. aureus* and *S. Enteritidis* human isolates from our study proved **susceptible** to the crude garlic extract indicating its therapeutic potential

Average **inhibition zones were slightly higher for *S. aureus* isolates**, which also exhibited reciprocal differences in susceptibility levels

Correlation between crude garlic extract concentrations and diameter of inhibition zones was observed.

Thank you for your attention

