ISSN (print ):2218-0230, ISSN (online): 2412-3986, DOI: http://dx.doi.org/10.21271/zjpas

### **RESEARCH PAPER**

# Isolation and identification of some Bacterial Species from Common carp (*Cyprinus carpio* Linnaeus, 1758) in Taqtaq District in Erbil Province, Kurdistan Region, Iraq.

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### ABSTRACT:

A number of 220 common carp *Cyprinus carpio* Linnaeus 1758, were collected from two ponds (98 from pond A and 122 from pond B) in Taqtaq District, northeast from Erbil Province, Kurdistan Region, Iraq. During September 2021 to April 2022. The fishes were examined for bacteria that infect them. In this research revealed the fortune of thirteen species of bacteria. The identification of the isolates was carried out depending on the morphology of the colony, specific media, and identification using VITEK II compact system (Biomerieux- USA). Recording of eight species of bacteria (*Streptpcoccus equi zooepidemicus, Methylobacterium spp, Rhizobium radiobacter, Burkholderia cepacia, B. multivorance, B. stabilis, B. vietnamiensis* and *Sphingobacterium thalpophilum*) are considered the first record in Iraq, and recording of five species of bacteria (*Staphylococcus lentus, Streptococcus thoraltensis, Klebsiella oxytoca, Pseudomonas aeruginosa* and *Serratia fonticola*) are the first record in Kurdistan Region.

KEY WORDS: *Cyprinus carpio*, Bacteria, Taqtaq ponds, Kurdistan Region, Iraq. DOI: <u>http://dx.doi.org/10.21271/ZJPAS.34.5.12</u> ZJPAS (2022) , 34(5);131-140 .

### 1. INTRODUCTION:

The beginnings of aquaculture in Iraq may be traced back to the mid-to-late twentieth century, whenever the common carp, *Cyprinus carpio* Linnaeus, 1758, had first been introduced in 1955 into the Al-Zaafaraniya fish farm in Baghdad City (Mhaisen, 1993). However, the culture sector in Kurdistan Region has only recently grown, and it has recently extended to several farms around the region (Mama and Abdullah, 2013).

*C. carpio* belongs to the Cyprinidae family, the biggest family of freshwater fishes in Iraq. The common carp is the most successful fish adapted in Iraq, whether in fish farms or most inland water bodies. *C. carpio* has a very important value among the fish species living in inland waters of the country

as regarding to its economic value and breeding features, and it has an important place between growth of the organism because of its omnivorous nature, rapid growth, ease of keeping in closed regions, viability in a wider range of water temperatures 3-35°C, considerable tolerance against a lack of oxygen, disease resistance, high fertility, easy reproduction, and they reach sexual maturity at 3-4 years of age, and it is comparatively delicious meat. Due to the common carp's adaptation to a wide range of climatic and geographical condition (Mama and Abdullah, 2012, Mustafa, 2016). All organisms, including common carp fish in the wild and on farms, are susceptible to the diseases (Price and Tom, 2006). Infectious diseases caused bv pathogenic organisms in the environment are classified as infectious diseases, while non-infectious diseases are caused by environmental issues, poor nutrition, or hereditary anomalies (Hardiono and Yanuhar, 2021).

Pathogenic diseases are more common and broadly classified as bacterial diseases, and they are typically associated with high mortality and morbidity rates, as well as widespread negative effects on farmers, consumers, and the environment. The disease can be caused by an unbalanced interaction between fish as the host, water (as the environment), and disease-causing agents (pathogens). Unbalanced interactions lead to stress and make it easier for pathogens to enter the fish body (Hardiono and Yanuhar, 2021).

Recently, the new method (VITEK 2) system was introduced and it's widely used all over the world, in addition, various methods using for the purpose. This system automatically performs all of the steps required for identification and antimicrobial susceptibility testing after a primary inoculum has been prepared and standardized (Nonhoff et al., 2005).

Aim of this research isolation and identification of some bacterial species from *C. carpio* in some ponds in Taqtaq District in Kurdistan Region, Iraq. Morphologically and by using VITEK 2 system.

### 2. MATERIALS AND METHODS

A number of 220 common carp fish belonging to *C. carpio* were gathered from tow ponds (98 from pond A and 122 from pond B) in Taqtaq District, during the period from September 2021 and the end of April 2022. Taqtaq District is in the Northeast 90km far from Erbil Provenance, Kurdistan Region, Iraq.

Fish samples (100-3000 g) Fishermen used cast nets and gill nets to collect the fish, which were then transported to the laboratory alive in a cool box filled with pond water to the Microbiology Lab of the Department of Biology, College of Education, Salahaddin University-Erbil, for bacteria isolation and identification.

In this research bacterial isolations, specimens were taken from skin, gills (especially in lesion places), blood and internal organs of fishes using sterile swabs, Bunsen burners are ignited and the inoculating swab loop is used for culturing of samples. The lid of the Petri dish containing the nutrient agar media and is opened slightly. The tip of the inoculating swab loop is then rubbed gently on the surface of the desired body part of the fish then streaking in nutrient agar, blood agar, and MacConkey agar. Cultured media were incubated at 37°C for 24-48 hours at the inverted position; the bacterial colonies were examined for characterization and identification. Morphological characteristics such as optical characteristics, size, shape, color, edge, and elevation were examined and recorded. The bacterial colonies were then subjected to Gram staining reaction and motility test identification of bacterial isolates from common carp fishes by VITEK II compact system was performed in Erbil International Hospital (Floris et al., 2021).

VITEK 2 system allows kinetic analysis by reading each test every 15 min. The optical system combines multichannel fluorimeter and photometer readings to record fluorescence, turbidity, and colorimetric signals (Garcia-Garrote et al., 2000).

### 3. RESULTS AND DISCUSSION

### 3.1. Staphylococcus lentus (Kloos et al., 1967)

#### Schleifer et al., 1983

This bacterium was isolated from the caudal fin of *C. carpio* in the pond (A) with a prevalence 2.45% (Table 1) and Table (2) show the identification of this species by using VITEK 2.

**Description:** Appeared as grayish white, white or cream and occasionally yellow or orange in the nutrient agar. Within 24 hours in air at 34-37°C, colonies are opaque and 2–7 mm in diameter. Gram positive, non-motile, non-sporing cocci of varying size occurring singly (0.7-1.2 µm in diameter), in pairs and in irregular clusters.

This bacterium was recorded in Iraq for the first time by (Al-Shemmari 2017) on *C. carpio*, no further record was reported for *Staphylococcus lentus*. So this is first record of this bacteria from fishes in Kurdistan Region (Mahisen, 2022).

## **3.2.** *Streptococcus thoraltensis* Devriese et al., **1997**

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This bacterium was isolated from the gills and skin of *C. carpio* from pond (B) with a prevalence 1.02% (Table 1) and Table (3) show the identification of this species by using VITEK 2.

**Description:** Appeared as cream and occasionally white or yellow in the nutrient agar and growth at 37 °C, colonies are opaque 1-5 mm in diameter. Gram positive organisms with cocci chains, with individual cells within the range of around 1.0-1.5 mm in diameter.

*S. thoraltensis* has been recorded in Iraq for the first time from *C. carpio* by (Al-Haider et al., 2019). No further record was reported for *S. thoraltensis*. So, this is first record of this bacteria from fishes in Kurdistan Region (Mhaisen, 2022).

*Streptococcus thoraltensis* is a recently described species that has been isolated from several animal species and from human.

### 3.3. Streptococcus equi zooepidemicus (ex Frost and Englebrecht, 1936) Farrow and Collins, 1985

This bacterium was isolated from the blood and mouth of *C. carpio* in the pond (B) with a prevalence 3.06% (Table 1) and Table (4) show the identification of this species by using VITEK 2.

**Description:** Appeared as circular, and opaque colored in the nutrient agar, colonies typically measure 0.5-1.5 mm in diameter. A convex elevation and a smooth surface. Its optimum temperature range of growth is 37 degrees Celsius. Gram-positive, non-sporulating, and non-motile bacteria The cells, which typically form in clusters or long branches, are encapsulated in a capsular polysaccharide that included hyaluronic acid. According to Mhaisen (2022), the present finding of *S. equi zooepidemicus* from *C. carpio* represents the first record of this species in Iraq.

*S. equi zooepidemicus.* rarely causes meningitis in humans by contact with domestic animals or their unpasteurized products.

### 3.4. Methylobacterium spp. Patt et al., 1976

This bacterium was found from the skin of *C*. *carpio* in the pond (B) with a prevalence 2.04% (Table 1) and Table (5) show the identification of this species by using VITEK 2.

**Description:** Appeared as pink or coral pigmented colonies on different media, including nutrient agar within 24 hours of incubation at 37°C, colonies are 1-2 mm in diameter. Grambacteria from negative the genus Methylobacterium that are non-spore-forming, strictly aerobic, and motile. According to Mhaisen present finding (2022).the of Methylobacterium from C. carpio represents the first record of this species in Iraq.

### 3.5. *Rhizobium radiobacter* (Beijerinck and van Delden 1902) Young et al., 2001

This bacterium was discovered in blood., mouth and skin of *C. carpio* in the pond (A and B) with a prevalence 4.09% and 7.14% respectively (Table 1) and Table (6) show the identification of this species by using VITEK 2.

Description: On a nutrient agar plate, the colony morphology is convex, circular, and smooth, and its color changes from nonpigmented to light beige. Colonies with 1 mm diameter growth. At 30 °C, mucoid white colonies were identified on nutrient agar within 48 hours. Aerobic, non-spore forming, Gram-negative rod-shaped (0.6–1.0 m 1.5–3.0 m) bacterium with 1–6 peritrichous flagella.

According to Mhaisen (2022), the present finding of Rhizobium radiobacter from *C. carpio* represents the first record of this species in Iraq.

*R. radiobacter* has been recognized as opportunistic pathogen in humans (Sawhney et al., 2016).

### 3.6. *Burkholderia cepacia* (Palleroni and Holmes, 1981) Yabuuchi et al., 1993

This bacterium was found from gills of *C*. *carpio* from the pond (A) with a prevalence 3.27% (Table 1) and Table (7) show the identification of this species by using VITEK 2.

**Description:** Colonies appear to be smooth and somewhat elevated, color formation is based on natural pigment expression and colonies vary from grey to sage green, with the medium changing from orange to bright pink bacteria will form visible pinpoint colonies within 24 h. Gram negative bacteria cells are 0.5 to 1.0 mm wide and 5 mm in length. They are rod-shaped, free-living, motile They have been found to possess

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multitrichous polar flagella as well as pili used for attachment.

According to Mhaisen (2022), the present finding of *B. cepacia* from *C. carpio* represents the first record of this species in Iraq.

In the 1950s, it was identified as a human pathogen. It was first isolated in cystic fibrosis (CF) patients in 1977 under the name *Pseudomonas cepacia*. During the 1980s (Frost et al., 2019).

### 3.7. Burkholderia multivorans Vandamme et al., 1997

This bacterium was found from mouth and gills of *C. carpio* from the pond (A and B) with a prevalence 2.45% and 3.06% respectively (Table 1) and Table (8) show the identification of this species by using VITEK 2.

**Description:** Appeared as purple color with a dry and puckered appearance after 24 to 48 hours of incubation at 30°C. Bacteria with a diameter of 1.6-3.2 mm that are Gram negative, motile, and rod-shaped. They have multitrichous polar flagella as well as pili that are used for adhesion. According to Mhaisen (2022), the present finding of *B. multivorans* from *C. carpio* represents the first record of this species in Iraq.

### 3.8. Burkholderia stabilis Vandamme et al. 2000

This bacterium was found from the gills of *C*. *carpio* in the pond (A and B) with a prevalence 1.63% and 6.12% respectively (Table 1) and Table (9) show the identification of this species by using VITEK 2.

**Description:** Colonies no pigmented strains have been detected and no melanin-like pigment is produced on tyrosine agar. Growing ability on nutrient agar. Gram-negative bacteria, cells are motile rods that are 1.0 to 2.0  $\mu$ m long and 0.6 to 0.9  $\mu$ m wide., after an incubation period of 48h at 37°C.

According to Mhaisen (2022), the present finding of *B. stabilis* from *C. carpio* represents the first record in Iraq.

**3.9.** *Burkholderia vietnamiensis* Gillis et al. 1995 This bacterium was found from the mouth of *C. carpio* in the pond (B) with a prevalence 5.10% (Table 1) and Table (10) show the identification of this species by using VITEK 2.

**Description:** Nutrient agar colonies are not pigmented. 0.8 to 2 mm long and 0.3 to 0.8 mm

wide motile cells Gram-negative bacteria colonies on king B medium do not produce a fluorescent pigment and are straight rods with a single polar flagellum or a tuft of polar. Growth occurs on nutrient agar at temperatures ranging from 20 to 41°C.

According to Mhaisen (2022), the present finding of *B. vietnamiensis* from *C. carpio* This is Iraq's first record of this species. In addition to the four *Burkholderia* species identified in this study (*B. cepacia, B. multivorans, B. stabilis, and B. vietnamiensi*). Another species of this genus, *B. pseudomonallei* from *C. carpio*, was found in Iraqi fishes Al-Shemmari (2017).

Some species of *B. vietnamiensis* are pathogenic for humans, animals and plants. Bacteria can cause divesting infections in patients with cystic fibrosis (CF) and chronic granulomatous disease (Gillis et al., 1995).

### 3.10. Klebsiella oxytoca (Flügge, 1886) Lautrop, 1956

This bacterium was found from the intestine, fin, skin, tail and gills of *C. carpio* in the pond (A and B) with a prevalence 4.09% and 8.16% respectively (Table 1) and Table (11) show the identification of this species by using VITEK 2.

**Description:** Colonies appear large, mucoid, and white in color. Gram-negative with a cylindrical rod shape measuring 2-5  $\mu$ m, straight shaped bacterium, and non-motile, capsulated, 0.3-6  $\mu$ m in diameter. is grows well on ordinary media to an optimal temperature of 37°C in 18-24 hours.

*K. oxytoca* was recorded from eight species of Iraqi fishes including *C. carpio* in south of Iraq. Therefore, could be considered this recording the first for *K. oxytoca* in Kurdistan Region (Mhaisen, 2022).

*Klebsiella oxytoca*, are considered as an indicator for sewage pollution and has been reported as opportunistic pathogen in fish (Leitner et al., 2015).

### 3.11. Pseudomonas aeruginosa (Schroeter, 1872) Migula,1900

This bacterium was isolated from the intestine of *C. carpio* in the pond (A) with a prevalence 0.81% (Table 1) and Table (12) show the identification of this species by using VITEK 2.

**Description:** In mixed cultures, it appears as a blue-green color and can be isolated as clear colonies on nutrient agar and grown at 42 °C. This blue green pigment is a combination of two *P*.

*aeruginosa* metabolites, pyocyanin (blue) and pyoverdine (green), which give cultures their characteristic blue-green color. Gramnegative, rod-shaped bacterium about 1-5mm long and 0.5-1.0mm wide encapsulated. *P. aeruginosa* produces colonies with a characteristic "grapelike" or "fresh-tortilla" odor on bacteriological media.

Previously, this species reported in Iraq from the same host by (Al-Faragi and Al-Saphar, 2012). It was then reported by two different fish hosts. Because there has been no previous report of this bacterium being recorded in the Kurdistan Region, this is the first record of *P. aeruginosa* in this region (Mhasein, 2022).

*Pseudomonas* are common inhabitants of soil, fresh water, and marine environments. *Pseudomonas aeruginosa* receives more attention since it is also an opportunist pathogen, causing human diseases (Michel-Briand and Baysse, 2002).

### 3.12. Serratia fonticola Gavini et al., 1979

This bacterium was found from the skin of *C*. *carpio* in the pond (B) with a prevalence 1.02% (Table 1) and Table (13) show the identification of this species by using VITEK 2.

**Description:** Appeared as red pigment, prodigiosin. Gramnegative, facultative anaerobic, rod in shape, and motile. They are usually 1–5 m long and spores aren't produced.

This species was isolated in Iraq for the first time from four different Marian fish species (Majeed et al., 2016). It was later recorded from *C. carpio* (Jassim et al, 2019). No further record was reported for *S. fonticola*. So, this is first record of this bacteria from fishes in Kurdistan Region (Mhaisen, 2022).

*S. fonticola* Water, soil, plants, and animals all contain it. *Serratia* was thought to be a harmless environmental bacterium until it was discovered that *S. marcescens*, the most common species in the genus, is an opportunistic pathogen of many animals, including humans. *S. marcescens* is most commonly associated with nosocomial (hospital-

acquired) infections in humans, but it can also cause urinary tract infections, pneumonia, and endocarditis (Singh et al., 1997).

### **3.13.** *Sphingobacterium thalpophilum* (Holmes et al., 1983) Takeuchi and Yokota, 1993

This bacterium was found from mouth of *C*. *carpio* from the pond (B) with a prevalence 1.02% (Table 1) and Table (14) show the identification of this species by using VITEK 2.

Description: Appeared as yellow or creamy white, no fluorescent pigment is produced. Gram negative, non-sporing rods 0.4-0.5 X 0.5-1.0tim. Non motile, non-gliding (no sliding translocation occurs). Aerobic, circular, entire, low convex, smooth and opaque, on nutrient agar grows at 37°C.

According to Mhaisen (2022), the present finding of *S. thalpophilum* from *C. carpio* represents the first record of this species in Iraq.

Finally, depending on the morphology of the bacterial colony and other methods are not accurate when compared with advanced technology (VITEK 2). In a traditional laboratory, using automated technology allows for easier, faster, and more reliable bacterial identification, which is necessary in medical cases.

### 4. CONCLUSIONS

Based on the results of research that has been done, the bacterial species isolated and identified from samples of carp, thirteen bacterial isolated, eight bacterial first new record in Iraq (Streptococcus equi zooepidemicus, Methylobacterium spp, Rhizobium radiobacter, Burkholderia cepacia, Burkholderia multivorans, Burkholderia stabilis, Burkholderia vietnamiensis and Sphingobacterium thalpophilum) and five bacterial first new record in Kurdistan Region (Staphylococcus Streptococcus lentus, thoraltensis, Klebsiella oxytoca, Pseudomonas aeruginosa and Serratia fonticola).

Table 1: The distribution of bacteria species on different sites of C. carpio in present study.

Species of Bacteria	Pond A (122 Fish)		Pond B (98 Fish)		Site of infection
	Number of	Prevalence	Number	Prevalence	
	infected fish	%	of infected fish	%	
Staphylococcus lentus	3	2.45	0	0	Tail
Streptococcus thoraltensis	0	0	1	1.02	Gills and Skin
Streptococcus equi spp zooepidemicus	0	0	3	3.06	Blood and Mouth
Methylobacterium spp	0	0	1	0.81	Skin
Rhizobium radiobacter	5	4.09	7	7.14	Blood, Mouth, Skin
Burkholderia cepacia	4	3.27	0	0	Gills
Burkholderia multivorans	3	2.45	3	3.06	Mouth and Gills
Burkholderia stabilis	2	1.63	6	6.12	Gills
Burkholderia vietnamiensis	0	0	5	5.10	Mouth
Klebsiella oxytoca	5	4.09	8	6.12	Intestine, Fin, Skin, Tail, Gills
Pseudomonas aeruginosa	1	0.81	0	0	Intestin
Serratia fonticola	0	0	1	1.02	Skin
Sphingobacterium thalpophilum	0	0	1	1.02	Mouth

	12 Sec. 2010											10					
Bic	chemica	al De	etail	s													
2	AMY	+	4	PIPLC	-	5	dXYL	+	8	ADH1	-	9	BGAL	-	11	AGLU	-
13	APPA	-	14	CDEX	+	15	AspA	-	16	BGAR	-	17	AMAN	-	19	PHOS	-
20	LeuA	-	23	ProA	-	24	BGURr	-	25	AGAL	-	26	PyrA	-	27	BGUR	-
28	AlaA	+	29	TyrA	-	30	dSOR	+	31	URE	-	32	POLYB	-	37	dGAL	+
38	dRIB	-	39	ILATk	-	42	LAC	+	44	NAG	+	45	dMAL	+	46	BACI	-
47	NOVO	-	50	NC6.5	-	52	dMAN	+	53	dMNE	+	54	MBdG	+	56	PUL	+
57	dRAF	+	58	0129R	-	59	SAL	+	60	SAC	+	62	dTRE	+	63	ADH2s	-
64	ОРТО	-							0.000								

Table 2: Species identification of staphylococcus lentus isolated by the VITEK 2.

Table 3: Species identification of streptococcus thoraltensis isolated by the VITEK 2.

Bio	chemica	I De	etail	S													
2	AMY	-	4	PIPLC	-	5	dXYL	+	8	ADH1	+	9	BGAL	-	11	AGLU	Τ-
13	APPA	-	14	CDEX	-	15	AspA	-	16	BGAR	-	17	AMAN	-	19	PHOS	ŀ
20	LeuA	+	23	ProA	-	24	BGURr	-	25	AGAL	+	26	PyrA	-	27	BGUR	- 1
28	AlaA	+	29	TyrA	-	30	dSOR	+	31	URE	+	32	POLYB	-	37	dGAL	+
38	dRiB	-	39	ILATk	-	42	LAC		44	NAG	+	45	dMAL	+	46	BACI	+
47	NOVO	+	50	NC6.5	-	52	dMAN	+	53	dMNE	+	54	MBdG	+	56	PUL	+
57	dRAF	+	58	0129R	-	59	SAL	+	60	SAC	+	62	dTRE	+	63	ADH2s	
64	OPTO	+															

Table 4: Species identification of streptococcus equi zooepidemicus isolated by the VITEK 2.

Bio	chemical	De	etail	S													
2	AMY	-	4	PIPLC	-	5	dXYL	-	8	ADH1	-	9	BGAL	-	11	AGLU	
13	APPA	-	14	CDEX	+	15	AspA	-	16	BGAR		17	AMAN	-2	19	PHOS	-
20	LeuA	+	23	ProA	-	24	BGURr	-	25	AGAL	-	26	PyrA	-	27	BGUR	-
28	AlaA	-	29	TyrA	-	30	dSOR	+	31	URE	+	32	POLYB	-	37	dGAL	
38	dRIB	-	39	ILATk	-	42	LAC	-	44	NAG	-	45	dMAL	-	46	BACI	-
47	NOVO	-	50	NC6.5	-	52	dMAN	-	53	dMNE	-	54	MBdG	-	56	PUL	+
57	dRAF	-	58	0129R	-	59	SAL	+	60	SAC	-	62	dTRE	-	63	ADH2s	-1
64	OPTO	-															

Table 5: Species identification of *Methylobacterium spp* isolated by the VITEK 2.

Bio	chemical	Det	tails									- <u>·</u>					•••••
2	APPA	-	3	ADO	+	4	PyrA	-	5	IARL	+	7	dCEL	-	9	BGAL	-
10	H2S	-	11	BNAG	-	12	AGLTp	-	13	dGLU	-	14	GGT	-	15	OFF	-
17	BGLU	-	18	dMAL	-	19	dMAN	+	20	dMNE	-	21	BXYL	-	22	BAlap	
23	ProA	-	26	LIP	-	27	PLE	+	29	TyrA	-	31	URE	+	32	dSOR	+
33	SAC	-	34	dTAG	+	35	dTRE	-	36	CIT	-	37	MNT	1-	39	5KG	
40	ILATk	-	41	AGLU	-	42	SUCT	-	43	NAGA	-	44	AGAL	-	45	PHOS	
46	GlyA	-	47	ODC	-	48	LDC	-	53	IHISa	-	56	СМТ	-	57	BGUR	-
58	0129R	-	59	GGAA	-	61	IMLTa	-	62	ELLM	-	64	ILATa	-			

Bic	chemica	l De	tails														
2	APPA	-	3	ADO	+	4	PyrA	-	5	IARL	+	7	dCEL	+	9	BGAL	-
10	H2S	-	11	BNAG	-	12	AGLTp	-	13	dGLU	-	14	GGT	-	15	OFF	-
17	BGLU	-	18	dMAL	+	19	dMAN	+	20	dMNE	+	21	BXYL	-	22	BAlap	-
23	ProA	-	26	LIP	-	27	PLE	+	29	TyrA	-	31	URE	-	32	dSOR	+
33	SAC	+	34	dTAG	+	35	dTRE	+	36	CIT	-	37	MNT	-	39	5KG	-
40	ILATk	-	41	AGLU	-	42	SUCT	-	43	NAGA	-	44	AGAL	-	45	PHOS	-
46	GlyA	-	47	ODC	-	48	LDC	-	53	IHISa	-	56	CMT	-	57	BGUR	-
58	0129R	-	59	GGAA	-8	61	IMLTa	-	62	ELLM	-	64	ILATa	-			

Table 6: Species identification of *Rhizobium radiobacter* isolated by the VITEK 2.

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Table 7: Species identification of Burkholderia cepacia isolated by the VITEK 2.

Bio	chemical	Det	ails				•										
2	APPA	-	3	ADO	+	4	PyrA	-	5	IARL	+	7	dCEL	+	9	BGAL	-
10	H2S	-	11	BNAG	-	12	AGLTp	-	13	dGLU	+	14	GGT	-	15	OFF	-
17	BGLU	-	18	dMAL	+	19	dMAN	+	20	dMNE	+	21	BXYL	-	22	BAlap	-
23	ProA	-	26	LIP	+	27	PLE	+	29	TyrA	-	31	URE	-	32	dSOR	+
33	SAC	+	34	dTAG	+	35	dTRE	+	36	CIT	-	37	MNT	-	39	5KG	-
40	ILATk	-	41	AGLU	-	42	SUCT	-	43	NAGA	-	44	AGAL	-	45	PHOS	-
46	GlyA	-	47	ODC	-	48	LDC	-	53	iHISa	-	56	CMT	-	57	BGUR	-
58	0129R	-	59	GGAA		61	IMLTa	-	62	ELLM	-	64	ILATa	-			

Table 8: Species identification of Burkholderia multivorans isolated by the VITEK 2.

Bio	chemical [	Det	ails														
2	APPA	-	3	ADO	+	4	PyrA	-	5	IARL	+	7	dCEL	+	9	BGAL	-
10	H2S	-	11	BNAG	-	12	AGLTp	-	13	dGLU	+	14	GGT	-	15	OFF	-
17	BGLU	-	18	dMAL	+	19	dMAN	+	20	dMNE	+	21	BXYL	-	22	BAlap	-
23	ProA	-	26	LIP	+	27	PLE	+	29	TyrA	-	31	URE	-	32	dSOR	+
33	SAC	+	34	dTAG	+	35	dTRE	+	36	CIT	-	37	MNT	-	39	5KG	-
40	ILATk	-	41	AGLU	-	42	SUCT		43	NAGA	-	44	AGAL	-	45	PHOS	
46	GlyA	-	47	ODC	-	48	LDC	-	53	IHISa	-	56	CMT	-	57	BGUR	-
58	0129R	-	59	GGAA	-	61	IMLTa	-	62	ELLM	-	64	ILATa	-			

Table	9:	Species	identificatoion	of	Burkholderia	stabilis	isolated	by	the	VITEK	2
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Bio	chemical [	Det	ails														
2	APPA	-	3	ADO	+	4	PyrA	-	5	IARL	+	7	dCEL	+	9	BGAL	-
10	H2S	-	11	BNAG	-	12	AGLTp	-	13	dGLU	+	14	GGT	-	15	OFF	-
17	BGLU	-	18	dMAL	+	19	dMAN	+	20	dMNE	+	21	BXYL	-	22	BAlap	-
23	ProA	-	26	LIP	+	27	PLE	+	29	TyrA	-	31	URE	-	32	dSOR	+
33	SAC	+	34	dTAG	+	35	dTRE	+	36	CIT	-	37	MNT	-	39	5KG	-
40	ILATk	-	41	AGLU	-	42	SUCT	-	43	NAGA	-	44	AGAL	-	45	PHOS	-
46	GlyA	-	47	ODC	-	48	LDC	1	53	IHISa	-	56	CMT	-	57	BGUR	-
58	0129R	-	59	GGAA	-	61	IMLTa	-	62	ELLM	-	64	ILATa	-			

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Bio	chemical l	Det	ails														100
2	APPA	-	3	ADO	+	4	PyrA	-	5	IARL	+	7	dCEL	+	9	BGAL	-
10	H2S	-	11	BNAG	-	12	AGLTp	-	13	dGLU	+	14	GGT	-	15	OFF	-
17	BGLU	-	18	dMAL	+	19	dMAN	+	20	dMNE	+	21	BXYL	-	22	BAlap	-
23	ProA	-	26	LIP	+	27	PLE	+	29	TyrA	-	31	URE	-	32	dSOR	+
33	SAC	+	34	dTAG	+	35	dTRE	+	36	CIT	-	37	MNT	-	39	5KG	-
40	ILATk	-	41	AGLU	-	42	SUCT	-	43	NAGA	-	44	AGAL	-	45	PHOS	-
46	GlyA	-	47	ODC	-	48	LDC	-	53	IHISa	-	56	СМТ	-	57	BGUR	-
58	0129R	-	59	GGAA	-	61	IMLTa	-	62	ELLM	-	64	ILATa	-			T

Table 10: Species identification of Burkholderia vietnamiensis isolated by the VITEK 2.

Bio	Biochemical Details																
2	APPA	-	3	ADO	+	4	PyrA	-	5	IARL	+	7	dCEL	-	9	BGAL	-
10	H2S	-	11	BNAG	-	12	AGLTp	-	13	dGLU	+	14	GGT	-	15	OFF	+
17	BGLU	+	18	dMAL	+	19	dMAN	+	20	dMNE	+	21	BXYL	-	22	BAlap	-
23	ProA	-	26	LIP	-	27	PLE	+	29	TyrA	-	31	URE	÷	32	dSOR	+
33	SAC	+	34	dTAG	+	35	dTRE	+	36	CIT	-	37	MNT	-	39	5KG	-
40	ILATk	-	41	AGLU		42	SUCT	-	43	NAGA	-	44	AGAL	-	45	PHOS	-
46	GlyA	-	47	ODC	-	48	LDC	-	53	IHISa	-	56	СМТ	-	57	BGUR	-
58	0129R	-	59	GGAA	-	61	IMLTa	-	62	ELLM	+	64	ILATa	-			

Table 12: Species identification of *Pseudomonas aeruginosa* isolated by the VITEK 2.

Bio	Biochemical Details																
2	APPA	-	3	ADO	+	4	PyrA	-	5	IARL	+	7	dCEL	-	9	BGAL	-
10	H2S	-	11	BNAG	-	12	AGLTp	-	13	dGLU	+	14	GGT	-	15	OFF	+
17	BGLU	-	18	dMAL	+	19	dMAN	+	20	dMNE	+	21	BXYL	-	22	BAlap	-
23	ProA	-	26	LIP	-	27	PLE	+	29	TyrA	-	31	URE	+	32	dSOR	+
33	SAC	+	34	dTAG	+	35	dTRE	+	36	CIT	-	37	MNT	-	39	5KG	-
40	ILATK	-	41	AGLU	-	42	SUCT	-	43	NAGA	-	44	AGAL		45	PHOS	-
46	GlvA	-	47	ODC	-	48	LDC	-	53	IHISa	-	56	CMT	-	57	BGUR	-
58	0129R	-	59	GGAA	-	61	IMLTa	-	62	ELLM	-	64	ILATa	-			

Table 13: Species identification of Serratia fonticola isolated by the VITEK 2.

Bio	Biochemical Details																
2	APPA	-	3	ADO	+	4	PyrA	-	5	IARL	+	7	dCEL	-	9	BGAL	+
10	H2S	-	11	BNAG	-	12	AGLTp	-	13	dGLU	+	14	GGT	-	15	OFF	+
17	BGLU	-	18	dMAL	+	19	dMAN	+	20	dMNE	+	21	BXYL	-	22	BAlap	-
23	ProA	-	26	LIP	-	27	PLE	+	29	TyrA	-	31	URE	+	32	dSOR	+
33	SAC	-	34	dTAG	+	35	dTRE	+	36	CIT	-	37	MNT	-	39	5KG	-
40	ILATk	+	41	AGLU	-	42	SUCT	-	43	NAGA	-	44	AGAL	-	45	PHOS	-
46	GlyA	-	47	ODC	+	48	LDC	+	53	IHISa	-	56	CMT	+	57	BGUR	+
58	0129R	+	59	GGAA	-	61	IMLTa	-	62	ELLM	+	64	ILATa	-1			

_	and an and the second sec																
Bio	3iochemical Details																
2	APPA	+	3	ADO	+	4	PyrA	-	5	IARL	+	7	dCEL	-	9	BGAL	-
10	H2S	-	11	BNAG	-	12	AGLTp	+	13	dGLU	-	14	GGT	-	15	OFF	
17	BGLU	+	18	dMAL	-	19	dMAN	-	20	dMNE	-	21	BXYL	-	22	BAlap	-
23	ProA	-	26	LIP	+	27	PLE	-	29	TyrA	+	31	URE	+	32	dSOR	+
33	SAC	-	34	dTAG	+	35	dTRE	-	36	CIT	-	37	MNT	-	39	5KG	- 1
40	ILATk	-	41	AGLU	+	42	SUCT	-	43	NAGA	-	44	AGAL	-	45	PHOS	-
46	GlyA	-	47	ODC	-	48	LDC	-	53	IHISa	-	56	CMT	-	57	BGUR	-
58	0129R	-	59	GGAA	+	61	IMLTa	-	62	ELLM	-	64	ILATa	-			

Table 14: Species identification of Sphingobacterium thalpophilum isolated by the VITEK 2.

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