2018

ABUNDANCE OF METHANOSAETA CONCILII-LIKE SPECIES IN THE SEDIMENT OF LEDBETTER CREEK EMBAYMENT OF KENTUCKY LAKE

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Recommended Citation

Von Dwingelo, Juanita, "ABUNDANCE OF METHANOSAETA CONCILII-LIKE SPECIES IN THE SEDIMENT OF LEDBETTER CREEK EMBAYMENT OF KENTUCKY LAKE" (2018). *Murray State Theses and Dissertations*. 86.
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ABUNDANCE OF *METHANOSAETA CONCILII*-LIKE SPECIES IN THE SEDIMENT OF LEDBETTER CREEK EMBAYMENT OF KENTUCKY LAKE

A Thesis
Presented to
The Faculty of the Department of Biology
Murray State University
Murray, Kentucky

In Partial Fulfillment
Of the Requirements for the Degree
Of Masters of Science

By Juanita Evelyn Von Dwingelo
April 2018

AKNOWLEGMENTS

I would first like to thank my mentor, Dr. Tim Johnston, you agreed to let me join your lab even though I'm sure I was a pain in Micro and I know I was a pain in Microbial Ecology! Without your help, guidance and support I would not be completing this degree (even though it took longer than either of us had anticipated) nor would I have gone on to continue and pursue a PhD. At least now you don't have to worry about me bothering you to get this done!

I would also like to thank my committee members who had no obligation to help me complete this degree after so many years. I very much appreciate your help and guidance. I have so many fond memories of Murray and many of them are thanks to you all.

Lastly, I would like to thank my parents and my husband – all of who encouraged me to see this through. I love you all and wouldn't be where I am today without you.

ABSTRACT

Reservoirs, including Kentucky Lake, are unique in that they do not follow a natural cycle of rising during the rainy seasons and dropping during the dry summers. The lake is manipulated, mainly for flood control and transportation, so that the water level is increased during the summer months and decreased during the winter months. This manipulation leaves the littoral sediment of the lake submerged in the summer and exposed in the winter. The seasonal water level, along with other factors, like farming practices in the watershed, contributes to the availability of nutrients for organisms inhabiting the sediment of the lake. However, the microbial communities of littoral zone sediments are not well understood. The Archaeal community of the Ledbetter Creek embayment, Kentucky Lake, has been the topic of study by several MSU students using sequence analysis of cloned 16S rDNA. BLAST searches showed that species related to the methanogen *Methanosaeta concilii* were the most abundant Archaea in this environment in 2007. Further, it was found that of the 59 *M. concilii*-like clones sequenced 50 were unique species (using 98.7% sequence identity as the cutoff). The aim of the present study was to again examine the Archaeal community in this environment to determine whether *Methanosaeta concilii*-like species continue to dominate the Archaeal community and to compare these sequences to those found in the previous study. DNA was extracted from sediment samples in 2011 & 2012; the 16S rDNA genes were amplified using Archaeal specific primers and cloned. The cloned inserts were sequenced and compared to the Genebank database and the *M. concilii*-like sequences found previously. The results from this study show that the *Methanosaeta concilii*-like species remain abundant in these samples, but are not the same species found before, using the 98.7% identity cutoff. The results of this study also found that there is another group of archaea that were abundant in this environment, *Methanoregula boonei*-like organisms. These data suggest that the Archaeal community is continually in flux in that the organisms are very closely related to those found in 2007 and may be derivatives, but the species change with time.

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Literature Review

ARCHAEA

Before the late 1970's evolutionary studies had been primarily confined to the metazoa and metaphyta, animals and plants, whose histories covered around 20% of the evolutionary time span. In 1866, Ernst Haeckel challenged the plant/animal division of the living world, recognizing that the protists did not fit into either category. He proposed that they had arisen separately from plants and animals. Haeckel's tree of life therefore had 3 main branches instead of only two. Later, in 1938, Herbert Copeland split the tree further creating a fourth branch, creating a kingdom for bacteria. In 1959, Robert Whittaker created a fifth branch to accommodate the fungi. Whittaker's scheme was the most widely accepted view of the organization of life, dividing the living world into five kingdoms – Plantae, Animalia, Fungi, Protista and Monera. Many however believed it to be incorrect phylogenetically. One such individual was Carl Woese [1]. In the 1970's using ribosomal RNA analysis, Woese and his collogues first recognized Archaea as one of the three major monophyletic lineages. Woese reclassified organisms into three domains instead of five kingdoms: Eucarya, which includes all eukaryotes, Bacteria, and Archaea [2]. This scheme is one that is most supported and accepted today.

Utilizing phylogenetic analysis, we can see relationships between these three domains. Phylogenetic trees (Fig. 1) based on rRNA sequences place Archaea and Eucarya on the same evolutionary branch, which suggests that Archaea, are more closely related to eukaryotes on a molecular level than they are to bacteria [1]. Archaea have some characteristics that are unique to them, and they have combinations of characteristics that were once thought to be unique to either the Bacteria or Eucarya. Members of the Archaea and Bacteria are united in the realm of prokaryotes by similarities in their cell size, lack of a nuclear membrane, and lack of organelles. Archaeal genes also appear to be organized into Bacteria-like operons and many of the archaeal operons are arranged in a fashion like the operons and gene clusters of the Bacteria [3, 4]. Another similarity between Archaea and Bacteria is that archaeal mRNAs lack 5'-end caps. Some have Shine-Dalgarno ribosome binding sites, but the locations of these Shine-Dalgarno sequences relative to the translational initiation codon are more variable in Archaea [5]. Several species of Archaea contain the bacterial cell division protein FtsZ, which is also thought to be a homolog of eukaryotic tubulins [6, 7]. Many members of Archaea contain a type II restriction enzyme system that is like those found in the Bacteria [8]. It was previously thought that both archaeal and bacterial protein-coding genes lack spliceosomal introns that are typically found in eukaryotic genes, but it has been shown that 16S and 23S rRNA of all Archaea, except *Euryarchaeota,* contain introns [9, 10]. Although the domains of Archaea and Bacteria seem to be very similar in some general genome organization, many of the archaeal genes show a greater similarity to eukaryotic homologs. Early studies using antibiotics showed genetic homology between the Archaea and the Eucarya [5]. For example, most bacteria are sensitive to streptomycin while Archaea and eukaryotes are both unresponsive to streptomycin. Archaea and eukaryotes are both sensitive to DNA polymerase inhibitors, like the antibiotic aphidicolin, while bacteria are resistant to aphidicolin [11]. There is evidence that shows significant similarities between Archaea and eukaryotic DNA replication, translational and transcriptional components. Archaeal DNA polymerases are homologous to eukaryotic DNA polymerases, neither of which are related to any bacterial DNA polymerases [12]. Evolutionarily, the RNA polymerases of Archaea are closer to the RNA polymerases of Eucarya. Phylogenetic trees constructed using the sequences for RNA polymerase subunits show that eukaryotic and archaebacterial genes are close relatives [13]. The numerous differences between Archaea, Bacteria and Eucarya listed above are what is responsible for making Archaea a unique organism.

Currently there are three potential scenarios for the evolution of the three domains of life. The first proposed scenario states that Bacteria diverged first from a lineage and produced Archaea and Eucarya. A second scenario states that a proto-eukaryotic lineage diverged from a prokaryotic lineage (Bacteria and Archaea). The third scenario, the one considered to be the most highly provisional, states that Archaea diverged first from a lineage leading to the eukaryotes and the Bacteria. Since the Archaea branch off closest to the root of the tree depicted in figure 1, the third scenario seems to make the most sense [14].

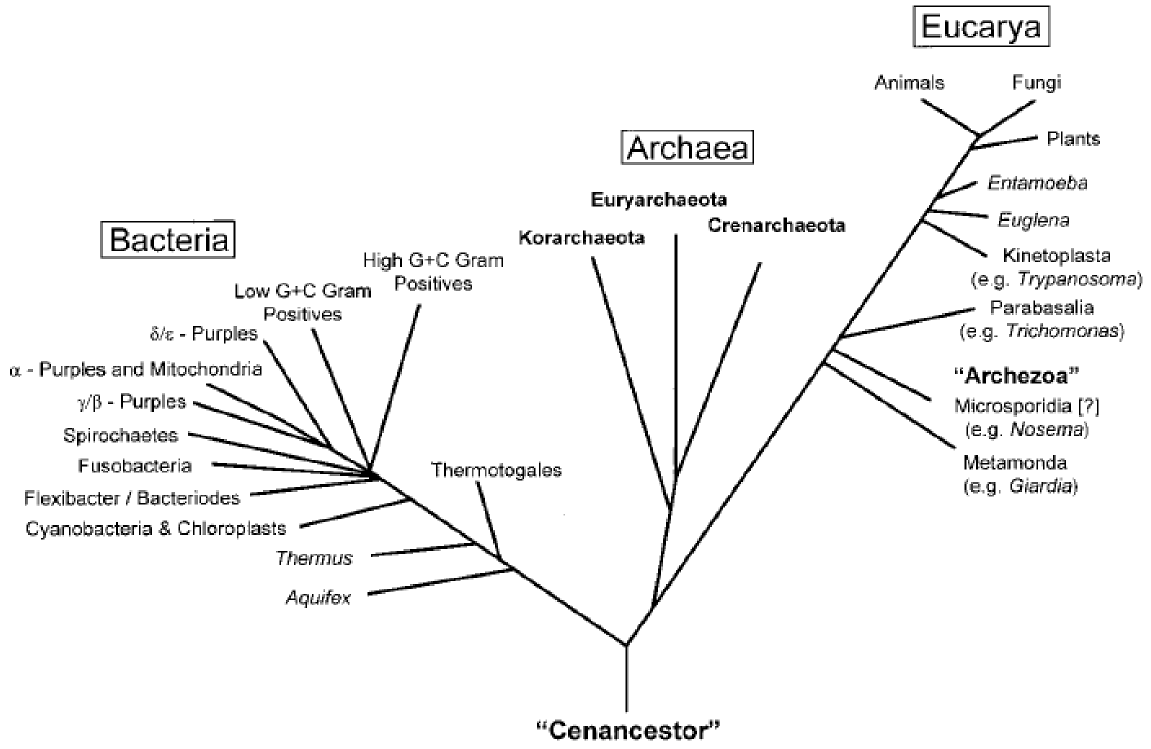


Figure 1: Universal Phylogenetic tree of the three domains of life based on rRNA gene sequencing (Brown 1997). Archaea diverged from a lineage producing Eucarya making them more closely related to the Eucarya and not the Bacteria.

Ecology

Archaea are known for their ability to live in environments that are considered uninhabitable and are known for their ability to dominate in these environments [15]. It was thought that Archaea only inhabited environments that were extreme: environments that had high salt concentrations, high and low pH, and high temperatures. Recent studies have shown that Archaea occur in many "mild" environments as well. These organisms exist in a wide variety of environments including ocean water, ocean sediment, gas hydrates (which is an ice-like mineral that crystallizes under conditions of low temperature, high pressure, and high gas concentrations), tidal flat sediments, freshwater lakes, soil, plant roots, petroleum-contaminated aquifers, the human mouth and gut, continental shelf anoxic sediments, in moderate-temperature hydrothermal vent microbial mats, in association with metazoan species, and in symbiotic relationships with marine plankton [16-18]. Archaea occupy a significant fraction of the total microbiota present, typically around 10% of the total rRNA phylotypes are Archaea, with the remainder composed of bacteria [16]. Although Archaea make up a significant portion of the microbes present, the ecological roles of many of these organisms is still unknown. Archaea, for the most part, can be phenotypically categorized into one of four groups: the methanogens, the extreme halophiles, thermoacidophiles, and the extreme thermophiles. Extremely halophilic archaea have been isolated from salterns, salt deposits, and landlocked seas. The methanogens are strictly anaerobic Archaea and have been isolated from diverse habitats including anoxic sediments, rice paddies, the rumen of cattle, the gut of termites, hydrothermal vents, and deep subterranean habitats. The thermophiles and thermoacidophiles are the heat and acid loving Archaea respectively. These organisms can be isolated from hydrothermal vents, hot springs, oil reservoirs, burning coal refuse piles and deep subterranean environments [2].

Taxonomy

Based on rRNA sequences, members of the domain Archaea phylogenetically fall into different distinct groups or kingdoms. It was previously thought that there were only two kingdoms, Crenarchaeota and Euryarchaeota, in which Archaea could fall, but in 1996 Barns, *et al*. proposed the existence of a third kingdom [19]. Their Ribosomal RNA studies showed that one group, pJP27/pJP78, did not associate with the two known kingdoms of Archaea phylogenetically, but they branched off below the Crenarchaeota-Euryarchaeota divergence. These organisms, which were isolated from a hot spring in Yellowstone National Park, were assigned their own kingdom, the Korarchaeota. Three kingdoms are currently recognized, but Crenarchaeota and Euryarchaeota are considered the two main kingdoms. The Euryarchaeota kingdom is phenotypically diverse and consists of the methanogens and their relatives. This kingdom is comprised of extreme halophiles, sulfate-reducing species, and two types of thermophiles [1]. The Crenarchaeota kingdom is not phenotypically diverse and consists mainly of thermophilic species such as the thermoacidophiles, sulfur-dependent archaebacteria, and extreme thermophiles. This kingdom is relatively homogeneous physiologically [1]. Although this group is mainly thermophilic there have been recent studies that have found the existence of non-thermophilic Crenarchaeota that have symbiotic relationships with plankton and that is associated with marine sponges and in sea ice. These Crenarchaeota thrive in frigid waters and sea ice, where they are present in significant numbers [20]. Because there are thermophilic species in both the Crenarchaeota and Euryarchaeota kingdom, thermophily is considered to be the ancestral phenotype of the Archaea [1].

The kingdom Crenarchaeota consists of only one class, the *Thermoprotei*. This class is subdivided into three orders: the *Thermoproteales*, the *Desulfurococcales*, and the *Sulfolobales*. As of 2001, when Bergeys Manual was published, there were around forty known species of Crenarchaeota. *Thermoproteales* consists of eight of the known species, all of which are rod shaped and are hyperthermophilic. There are two families in the *Thermoproteales* order, *Thermoproteaceae* and *Thermofilaceae*. The *Thermoproteaceae* family contains four genera. Genus I, *Thermoproteus*, contains three species. Genus II, *Caldivirga*, contains one species. Genus III, *Pyrobaculum*, contains three species. Genus IV, *Thermocladium,* contains one species. The *Thermofilaceae* family consists of one genera, *Thermofilum,* with no defined species. The *Thermoproteus* and *Thermofilum* genera are two of the most studied genera within this order; both genera have been isolated from neutral or slightly acidic hot springs. The second order, *Desulfurococcales*, consists of seventeen species and two families: *Desulfurococcaceae* and *Pyrodictiaceae*. The first family, *Desulfurococcaceae,* consists of eight genera. Genus I, *Desulfurococcus*, consists of four species. Genus II, *Aeropyrum*, consists of one species. Genus III, *Ignicoccus*, consists of two species. Genus IV, *Staphylothermus*, genus V, *Stetteria*, genus VI, *Sulfophobococcus,* genus VII*, Thermodiscus,* and genus VIII, *Thermosphaera*, all consist of one species. The second family, *Pyrodictiaceae*, consists of 3 genera. Genus I, *Pyrodictium*, consists of three species. Genus II, *Hyperthermus*, and genus III, *Pyrolobus* consist of only one specie each. Organisms belonging to the genus *Desulfurococcales* have been isolated from volcanic habitats, which include deep-sea hydrothermal vents, and shallow water thermal springs. The last order, *Sulfolobales*, consists of fifteen species, one family and 6 genera. Genus 1, *Sulfolobus*, consists of six species. Genus II, *Acidianus*, consists of three species. Genus III, *Metallosphaera*, consists of two species. Genus IV, *Stygiolobus*, and genus V, *Sulfurisphaera*, consists of one species. Lastly, genus VI, *Sulfurococcus*, consists of two species. *Sulfolobus* and *Acidianus* appear to be the two key genera within this family and have been isolated from volcanic habitats [21]. The Crenarchaeota that have been found to inhabit colder waters have not yet been cultured and what is known about them is information provided by 16S rRNA analysis [20].

The second kingdom of Archaea, Euryarchaeota, is much larger and more diverse than the Crenarchaeota kingdom. This kingdom currently consists of seven classes: the *Methanobacteria*, the *Methanococci*, the *Halobacteria*, the *Theroplasmata*, the *Thermococci*, the *Archaeoglobi*, and the *Methanopyri*. As of 2001 there were 138 species of Euryarchaeota that have been identified. Within the *Methanobacteria* class there are five orders: *Methanobacteriales*, *Methanococcales*, *Methanomicrobiales*, *Methanopyrales*, and *Methanosarcinales*. The order

Methanobacteriales consists of two families, *Methanobacteriaceae* and *Methanothermaceae*, and twenty-five species. The *Methanococci* class consists of three orders: *Methanococcales*, *Methanomicrobiales*, and *Methanosarcinales*. *Methanococcales* consists of two families, *Methanococcaceae* and *Methanocaldococcaceae*, and eight species. *Methanomicrobiales* consists of three families, *Methanomicrobiaceae*, *Methanocorpusculaceae* and *Methanospirillaceae,* and twenty-three species. *Methanosarcinales* consists of eighteen species and six families: *Methanosarcina*, *Methanococcoides*, *Methanohalobium*, *Methanohalophilus*, *Methanolobus*, and *Methanosalsum*. Organisms in the *Methanobacteriales*, *Methanococcales*, *Methanomicrobiales*, and *Methanosarcinales* orders are all obligate anaerobes that are capable of producing methane as part of their metabolism. As of 2001, there have been approximately 100 species of methanogens identified [21]. These organisms have been isolated from many different habitats including animal digestive tracts, hydrothermal vents, and anoxic sediments [20].

The Halobacteria class of Euryarchaeota contains a diverse group that inhabits environments that are high in salt. These are the extreme halophiles and haloalkaliphiles. In order for an organism to be considered an extreme halophile it must require at least 9% NaCl for growth. Most species of extreme halophiles require 12-23% NaCl for optimal grown and virtually all extreme halophiles can grow at a concentration of 32% NaCl [20]. Currently there are 35 species of Halobacteria that have been identified. This class consists of one order, Halobacteriales, which has one family, Halobacteriaceae. This family consists of fourteen genera: *Halobacterium, Haloarcula, Halobaculum, Halococcus, Haloferax,* *Halogeometricum, Halorubrum, Haloterrigena, Natrialba, Natrinema, Natronobacterium, Natronococcus, Natronomonas,* and *Natronorubrum* [21]. Species from this class have been isolated from environments that are naturally salty, such as solar salt evaporation ponds and salt lakes, and artificial saline habitats, such as the surfaces of foods that are heavily salted [20].

The Thermoplasmata class also consists of one order, *Thermoplasmatales*, and this order has two families, Thermoplasmataceae and Picrophilaceae. There are only three genera and four species that have been described for this class [20]. Organisms belonging to this class are among the most acidophilic of all known microorganisms, some being able to live at a pH below 0. These organisms have been isolated from environments that have low pH and high temperatures including coal refuse piles, mine tailings, and acidic Japanese solfataras [20]. The Thermococci class contains one order, *Thermococcales*, which contains one family, Thermococcaceae. There are only two genera known for this class. The first genus, *Thermococcus*, consists of fifteen known species. The second genus, *Pyrococcus*, consists of five species. Organisms belonging to this class are hyperthermophilic heterotrophs and most need sulfur for growth and require 75˚C to 100°C for optimal growth. These organisms have been isolated, mainly, from deep-sea and shallow marine hydrothermal vents and terrestrial thermal springs [21].

The sixth class, Archaeoglobi, consists of one order, *Archaeoglobales*, and one family, Archaeoglobaceae. There are two known genera in this class: *Archaeglobus,* which contains four species, and *Ferroglobus*, which contains only one species [21]. Organisms belonging to this class are hyperthermophilic and they catalyze anaerobic respiration where elemental sulfur is used as an electron acceptor causing it to be reduced. Species belonging to this class have been isolated from shallow marine hydrothermal vents, and hot marine sediments near hydrothermal vents [20]. The last class, Methanopyri, also consists of one order, *Methanopyrales*, and one family, Methanopyraceae. There is one genus, *Methanypyrus*, and one species known in this class [21]. *Methanopyrus* has been isolated from hot sediments near submarine hydrothermal vents and from the walls of black smoker hydrothermal chimneys. These organisms grow rapidly at temperatures greater than 100°C but will not grow at temperatures lower than 80°C [20].

Archaea have previously been divided into four groups, Group I- IV. Groups I and II appear to be the most abundant, widely distributed and ecologically diverse, which has made them the easiest to study [18]. Group I Archaea have been found in both terrestrial and marine environments. This group is related to Crenarchaeota and it is thought that they could comprise as much as 20% of total microbial biomass found in the world's oceans. Groups II and III are related to Euryarchaeota. Group III Euryarchaeota have been found in waters below the photic zone and are peripherally related to the order *Thermoplasmatales* [2]. They can also be found in marine sediments and marine plankton [18]. The Group IV Euryarchaeota inhabit deep ocean waters and are related to haloarchaea [2].

While there is a wide array of methane producing Archaea, it is important to note that methanogen is not a phylogenetic group or even a taxon. Methanogen is simply a description based on the end product of their metabolic processes.

METHANOGENS

Methanogenic Archaea is a diverse group of Euryarchaeota that are strictly anaerobic organisms with an energy metabolism that is restricted to the formation of methane from formate, methylamines, methanol, CO_2 and H_2, and/or acetate [22]. Studies show that methane is more than 20 times more effective as a greenhouse gas than carbon dioxide and methane can persist in the atmosphere for 9-12 years [23]. It has also been estimated that 1 billion tons of methane is formed globally per year by methanogenic Archaea in different environments. This means that 2% of the net CO_2 that is fixed annually into biomass by photosynthesis would end up as methane [22]. Around 70% of methane arises from four main sources: natural wetlands and tundra, rice paddies, ruminants, and fossil fuels [23]. Most of the 70% of methane emitted is derived from biological methanogenesis [24]. In the last 200 years, the atmospheric concentration of methane has increased threefold. With this increase in methane present in the environment, the study of methanogens has become increasingly important.

Methanogens have been cultivated from a wide variety of environments. They are common in environments with extreme temperatures, salinity and pH as well as temperate and other mild habitats. Methane producing Archeae are unique in that they are the only group of organisms that have species that are capable of growth at 0°C and others at 110°C [25]. Common methanogen habitats include marine sediments, freshwater sediments, flooded soils, human and animal gastrointestinal tracts, termites, anaerobic digesters, landfills, geothermal systems, and heartwood of trees [24]. Methanogenic Archaea are known as being slow growing and are strictly anaerobic, requiring nutrients and trace elements that make them hard to cultivate [26]. The use of 16S rRNA analysis overcomes this difficulty because it can be carried out without cultivation, making this method a powerful tool for the analysis of methanogens [27].

Morphology

Methane producing Archaea are diverse and their diversity is dramatically illustrated by their variety of morphological, physiological, and biochemical characteristics. Methanogens exhibit a wide range of cellular morphologies, including cocci, packets of cocci (referred to as pseudosarcina), rods of varying length and shape, and spirillum [28]. Some species exist in irregular, unusual flattened plates. Some species can aggregate in clusters. Motility is present in some. The gram reaction for methanogens can be either positive or negative even within members of the same genus [29]. The composition of the cell envelopes in methanogenic Archaea are very different from that of Bacteria and are diverse among the methanogens. Methanogen cell envelopes lack murein, which is the peptidoglycan found in Bacteria cell walls, instead they contain pseudomurein or protein subunits depending on the order. This lack of murein renders methanogens insensitive to the antibiotics like penicillin that inhibit the synthesis of cell walls in Bacteria. The lipids found in the cell envelopes are also unusual and the polar lipids of methanogens are unlike membrane glyco- and phospholipids of Bacteria.

Methanogenesis

Methanogens are a phylogenetically diverse group, but they are a rather uniform group of strictly anaerobic Archaea nutritionally. These organisms are able to grow by the conversion of certain compounds to methane. This simple pathway is coupled to the generation of ion gradients across the membrane that is used to drive the synthesis of ATP [30]. The methanogens are the only known archaebacteria that couple methane synthesis to energy generation [28]. Methanogenesis is really not a form of anaerobic respiration although it is a type of energy generating metabolism that needs an outside electron receptor in the form of CO_2. Although methanogens are similar nutritionally and all employ similar pathways, they differ significantly in regards to what compounds are involved in the proton motive electron transport chain, and because of this they most likely use different mechanisms to generate the proton gradient [30].

The 83 species of methanogenic Archaea that have been described as of 2000 have been divided into three classes, or main nutritional categories, based on the substrate used in methanogenesis (Fig. 2). The first nutritional category consists of 61 species of hydrogenotrophs, which oxidize H_2 and reduce CO_2 to form methane, and 38 species of formatotrophs, which oxidize formate to form methane. The second nutritional category consists of twenty species of methylotrophs, which use methyl compounds to make methane. The third nutritional category consists of nine species of acetotrophic methanogens, which use acetate to produce methane. There are some species of methanogens that share nutritional characteristics and therefore cannot be placed in one single group [29].

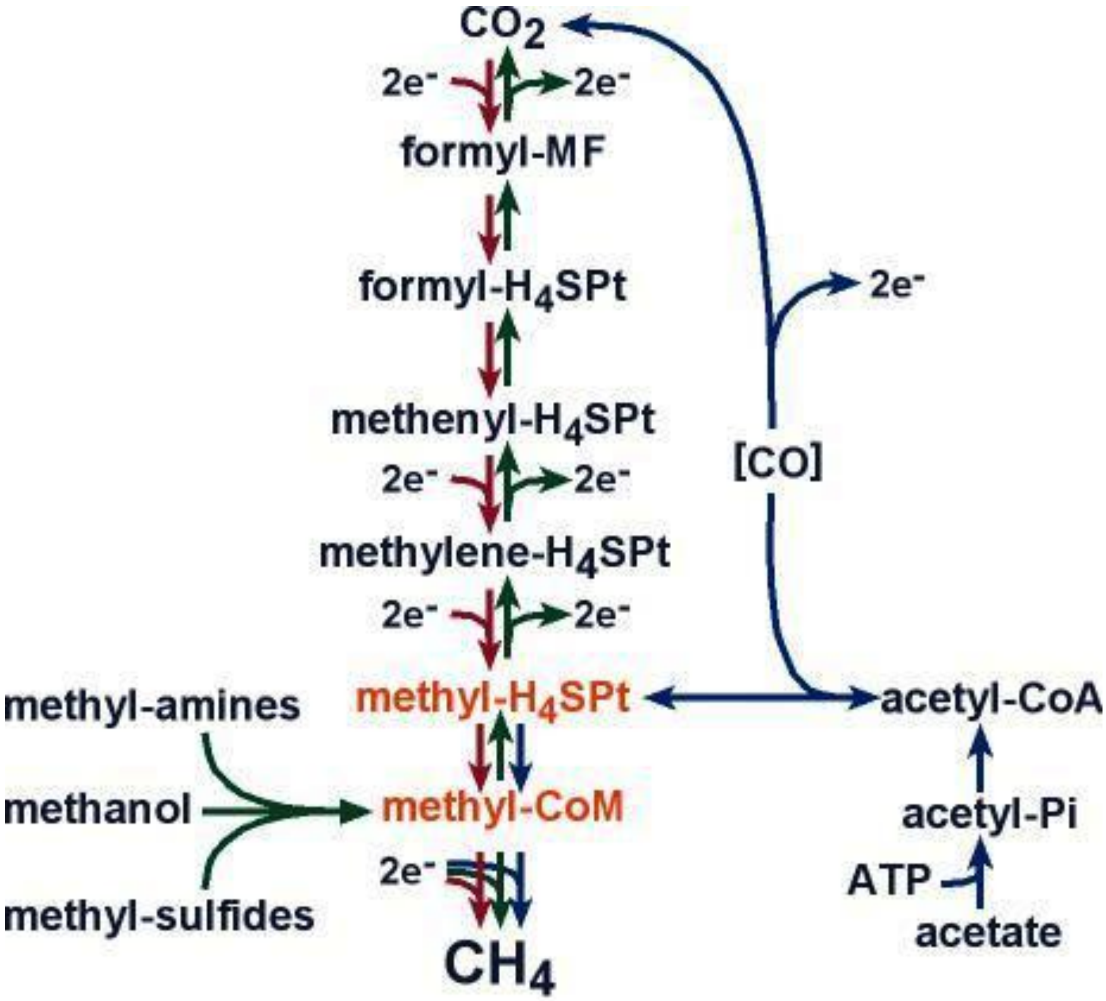


Figure 2: The three pathways of methanogenesis. The red arrows represent the hydrogenotrophic pathway. The green arrows represent the methylotrophic pathway and the blue arrows represent the acetoclastic pathway [24].

Methane can be produced via three different pathways (Fig. 2). These pathways differ in the substrates used. The three major substrates utilized are CO_2, methyl-group containing compounds, and acetate [24]. The first pathway is the hydrogenotrophic pathway. Most methanogens are hydrogenotrophs that can reduce CO_2 to methane, making this pathway the most widespread and it can be found in all methanogenic orders. This pathway involves the reduction of CO_2 with H_2 as an electron donor [31]. The second pathway is known as the methylotrophic pathway. This pathway has several variants; the best studied being the version where C-1 compounds such as methylamines or methanol can be used as both an electron donor and an electron acceptor [31].

The third and final pathway is the acetoclastic pathway. This pathway utilizes acetate, which is a major intermediate in the anaerobic food chain and as much as two-thirds of biologically produced methane is derived from acetate [24]. Based on free energy values for methanogenesis reactions, the most favorable reaction is the reduction of CO_2 by H_2 and the least favorable is the acetoclastic reaction (Table 1). This has led to a natural selection pressure that has resulted in the evolution of many more hydrogenotrophic species than acetotrophs. The large number of species within the first class of methanogens also illustrates this. Because there is no selection pressure in favor of acetotrophs, there is less evolution, which has led to a decrease in species diversification for this group. The low number of species within the third class of methanogens illustrates this [24].

Taxonomy

Table 1: The standard changes in free energies for methanogenesis reactions with species that use each reaction [24].

Reaction	$\Delta G^{\circ\prime\,a}$ (kJ/mol CH_4)	Organisms
I. CO_2-type		
$4\ H_2 + CO_2 \rightarrow CH_4 + 2\ H_2O$	−135	Most methanogens
$4\ HCOOH \rightarrow CH_4 + 3\ CO_2 + 2\ H_2O$	−130	Many hydrogenotrophic methanogens
CO_2 + 4 isopropanol → CH_4 + 4 acetone + $2\ H_2O$	−37	Some hydrogenotrophic methanogens
$4\ CO + 2H_2O \rightarrow CH_4 + 3\ CO_2$	−196	*Methanothermobacter* and *Methanosarcina*
II. Methylated C1 compounds		
$4\ CH_3OH \rightarrow 3\ CH_4 + CO_2 + 2\ H_2O$	−105	*Methanosarcina* and other methylotrophic methanogens
$CH_3OH + H_2 \rightarrow CH_4 + H_2O$	−113	*Methanomicrococcus blatticola* and *Methanosphaera*
$2\ (CH_3)_2\text{-}S + 2\ H_2O \rightarrow 3\ CH_4 + CO_2 + 2\ H_2S$	−49	Some methylotrophic methanogens
$4\ CH_3\text{-}NH_2 + 2\ H_2O \rightarrow 3\ CH_4 + CO_2 + 4\ NH_3$	−75	Some methylotrophic methanogens
$2\ (CH_3)_2\text{-}NH + 2\ H_2O \rightarrow 3\ CH_4 + CO_2 + 2\ NH_3$	−73	Some methylotrophic methanogens
$4\ (CH_3)_3\text{-}N + 6\ H_2O \rightarrow 9\ CH_4 + 3\ CO_2 + 4\ NH_3$	−74	Some methylotrophic methanogens
$4\ CH_3NH_3Cl + 2\ H_2O \rightarrow 3\ CH_4 + CO_2 + 4\ NH_4Cl$	−74	Some methylotrophic methanogens
III. Acetate		
$CH_3COOH \rightarrow CH_4 + CO_2$	−33	*Methanosarcina* and *Methanosaeta*

Phenotypic characteristics are often not sufficient in means of distinguishing taxa or determining the phylogenetic position of a taxon [32]. The methanogens were the first microbial group to have their taxonomy based on 16S rRNA sequence divergence [33]. Methanogens, classically, were grouped into one of three orders based on work done by Balch *et al*. in 1979 [32]. They had proposed a taxonomic revision of methanogens based on 16S rRNA oligonucleotides and placed the 13-available species at the time into three orders, four families, and seven genera. In 1988, Boone and Whitman proposed a minimal standard for describing new methanogenic taxa [33]. These minimal standards include morphology, Gram staining, culture purity, electron microscopy, motility, colony morphology, susceptibility to lyse, nutritional spectrum, growth rates, end products, antigenic fingerprinting, G+C content of the DNA, growth conditions, lipid analysis, nucleic acid hybridization, distribution of polyamines, 16S rRNA sequencing, and sequence analysis [33]. These criteria were approved and accepted by the Subcommittee for Taxonomy of Methanogens and based on these criteria, Boone *et al*. could define five orders consisting of ten families and 31 genera and (as of 2000) 74 validated species (Fig. 3) [29, 33]. The five orders are *Methanobacteriales, Methanococcales, Methanomicrobiales, Methanosarcinales*, and *Methanopyrales*, all of which belong to the kingdom Euryarchaeota and the phylum Euryarchaeota. The *Methanobacteriales* order encompasses the non-motile methanogens. Members of this order generally use CO_2 as the electron acceptor and H_2 as the electron donor to produce methane. This order is divided into two families, *Methanobacteriaceae* and *Methanothermaceae*. The family *Methanobacteriaceae* contains four genera:

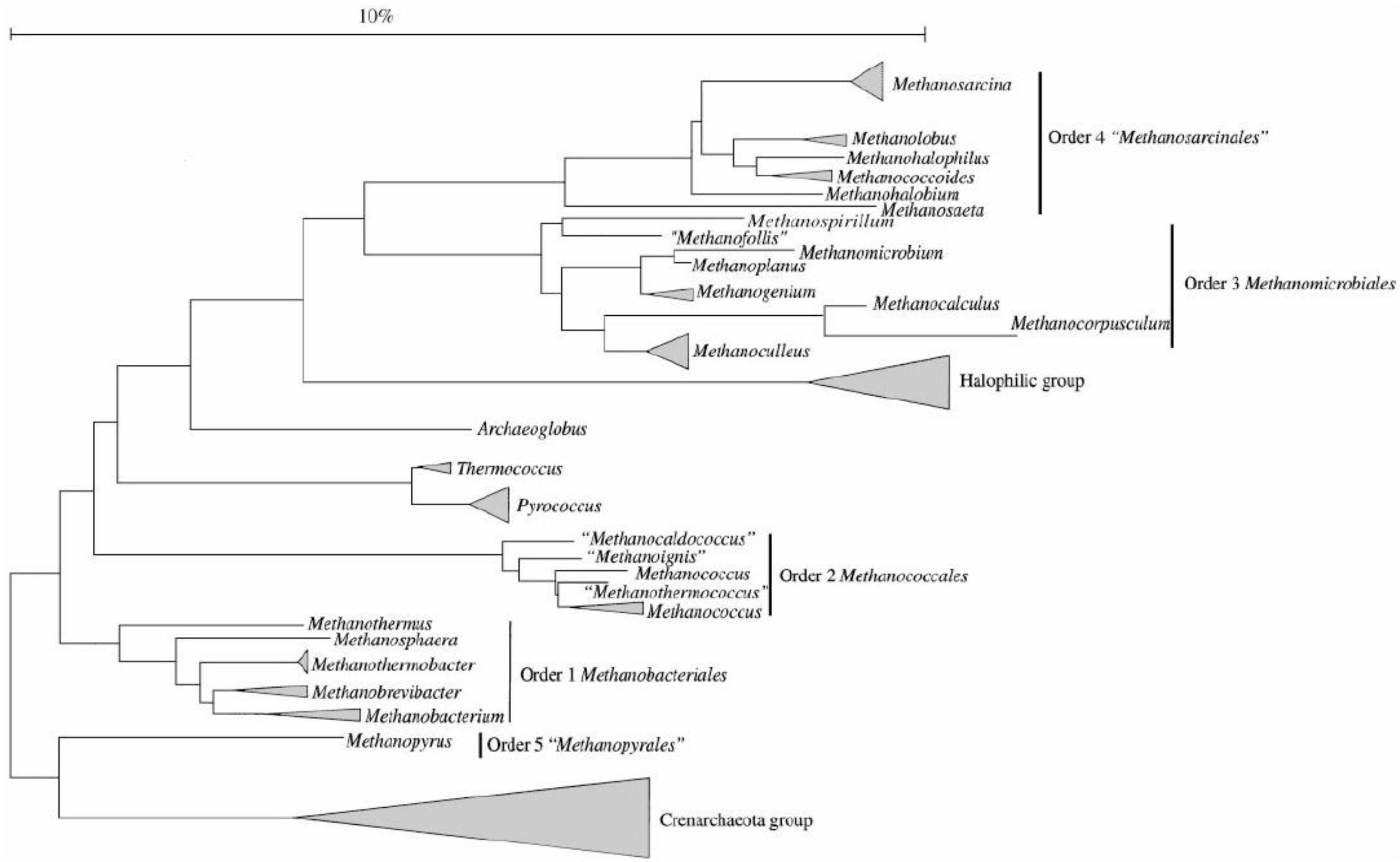


Figure 3: Phylogeny of methanogens, proposed by Boone *et al.* Large triangles represent non-methanogens which were used to help root the tree [35]. Figure shows the different orders of Methanogens and the members of each order.

Methanobacterium, Methanobrevibacter, Methanosphaera, and Methanothermobacter. The first genus, Methanobacterium, contains 13 species, all of which use H2 and CO2 for methanogenesis and six species can utilize formate. All cells within this genus are rod or filamentous in morphology. Some of the species are thermophilic and a few are acidophilic. These organisms can be found in freshwater habitats. The *Methanothermobacter* genus was created to include three species of thermophilic methanogens. *Methanobrevibacter* contains seven species that are neutrophilic mesophilic short rods, which often form pairs or chains. Each of the species in this genus inhabits a specialized habitat, including bovine rumen, sewage sludge, and intestinal tracts of human and animals. The last genus, *Methanosphaera,* contains two species, both of which are Gram-positive spherical-shaped organisms that are generally observed in the digestive tracts of animals and have been isolated from human and rabbit feces. The family *Methanothermaceae* contains only one genus, *Methanothermus*, and two species. Both species are extreme thermophiles and have been isolated from only a specific habitat, volcanic springs. These species have cells that are rod shaped and these cells contain a double-layered cell wall [29].

The order *Methanococcales* consists of two families and four genera. Members of this order are hydrogenotrophic methanogens that have been isolated from marine and coastal environments. All species in this order are irregular cocci and contain proteinaceous cell walls. Species are also motile by a polar tuft of flagella and cells lyse quickly in detergents. All species use both H_2 and formate as the electron donors for methanogenesis. The two families in this order are *Methanococcaceae* and *Methanocaldococcaceae.* The *Methanococcaceae* family is comprised of two genera. The *Methanococcus* genus includes five species, all of which are mesophilic. The *Methaothermococcus* genus includes one thermophilic species. The *Methanocaldococcaceae* family includes two thermophilic genera, each of which contains one species: *Methanocaldococcus jannaschii* and *Methanoignis igneus* [29].

The third order, *Methanomicrobiales*, is comprised of three families and nine genera of methanogens that are hydrogenotrophic. The first family is *Methanomicrobiaceae,* which contains seven of the genera. Members of this family have a variety of different morphologies including small rods, irregular cocci, and plane-shaped cells. The cell walls of members of this family are proteinaceous. Most of the strains in this family can utilize formate and some secondary alcohols for methanogenesis. The genus *Methanomicrobium* contains one mesophilic species, which has been isolated form bovine rumen. The genus *Methanolacinia* contains three reclassified species that have been isolated from marine sediments and are unable to use formate. The genus *Methanogenium* contains five species that are Gram-negative, non-motile, cocci. Species in this genus have been isolated from thermophilic species. The five-mesophilic species can use formate for methanogenesis. The genus *Methanoplanus* is comprised of three species of organisms that are plane-shaped and have a polar tuft of flagella. These organisms use formate for methanogenesis. One species in this genus is an endosymbiont of marine ciliates and has been found in close association with microbodies, which are thought to provide hydrogen to the methanogen. The genus *Methanofollis* contains two species, which can use formate to make methane. The last genus, *Methanocalculus*, is a newly described genus that consists of one species, *M. halotolerans*, an irregular coccoid that was isolated from an offshore oil well. This methanogen is a hydrogenotrophic halotolerant organism that grows optimally at 5% NaCl and will tolerate up to 12% NaCl. The second family in this order, *Methanocorpusculaceae*, contains one genus, *Methanocorpusculum*, and five species. All species are mesophilic, small coccoid methanogens with monotrichous flagella. These species use H_2/CO_2 and formate for methanogenesis and some species can use 2-propanol/CO_2. The last family, *Methanospirillaceae*, consists of one genus, *Methanospirillum*. Members of this genus have been isolated from various habitats and are mesophilic. Only one species in this genus has been described so far, *Methanospirillium hungatei*. This species uses H_2 and CO_2 as well as formate for methanogenesis. Some strains can use 2-propanol and 2-butanol along with CO_2 [29].

The fourth order, *Methanosarcinales*, was created to regroup all the acetotrophic and methylotrophic methanogens. This order consists of two families. The first family, *Methanosarcinaceae* consists of six genera and 21 species. The genus *Methanosarcina* is comprised of eight species of Gram-positive, non-motile, acetotrophic methanogens. This genus predominates in many anaerobic ecosystems where organic matter is completely degraded to CH_4 and CO_2 [29]. Members of this genus have been isolated from freshwater and marine mud, anoxic soils, animal-waste lagoons, and anaerobic digesters. Some species can use H_2 and

CO_2, acetate and methyl compounds for methanogenesis making these species versatile. All eight species exhibit a pseudo sarcina, or cube like, cell arrangement and morphology. The remaining five genera are methylotrophs, meaning they utilize only methanol and methylamine for methanogenesis. These genera are all non-motile, most are mesophilic and species have cell morphologies that are irregular coccoid. Most organisms in these genera have been isolated from high salt concentration environments. The genus *Methanolobus* consists of five species, which grow optimally when the NaCl concentration is around 0.5 M. The genus *Methanococcoides* has two species whose growth is optimal in NaCl concentrations of 0.2-0.6M and high concentrations of magnesium are required for growth. This genus includes the species *Methanococcoides burtonii* which was first isolated from Ace Lake in Antarctica and the optimum temperature for this organism is 23°C, but it survives at a range of 4°C-29°C. The genus *Methanosalsus* contains one species. This species is an alkaliphilic, halophilic species that was isolated from an Egyptian Lake. The last genus, *Methanohalobium*, consists of one species that is extremely halophilic and grows optimally at 25% NaCl and 50°C [29].

The last order, *Methanopyrales*, represents a novel group of methanogens which grow at 110°C and are unrelated to all other known methanogens. This order consists of one family, *Methanopyraceae*, which includes only one species, *Methanopyrus kandleri*. This species is a Gram-positive hydrogenotrophic, hyperthermophilic archaeum that has been isolated from hydrothermally heated deep-sea sediments as well as a shallow marine hydrothermal system [29].

Ecology

The distribution of methanogens in the environment is dependent on many different factors including temperature, pH and salinity. Most of the methanogens grow within a narrow pH range, from 6.0 to 8.0. There are some exceptions to this and there are a few species that can tolerate pH's outside of this range. Methanogens can grow at a much wider temperature range, which ranges from 0°C to 110°C. Tolerance to salt concentrations is variable depending on which species of methanogen is being considered. Marine methanogens tend to inhabit environments with higher salt concentrations since they tend to be more halophilic [29].

A variety of substances have been found that act as metabolic inhibitors for methanogens. These inhibitors play an important role ecologically because they can greatly reduce the distribution of methanogens. Many of the chemical substances are toxic and can physiologically block methanogenesis. Methanogenesis is sensitive to chlorinated methanes, ethylene, acetylene, monensin, and possibly heavy metals. Methanogens are also strict anaerobes and are very sensitive even to levels of oxygen less than 10 ppm. Levels of oxygen greater than 10 ppm can cause an irreversible dissociation of the F_{420}-hydrogenase enzyme complex. This result probably occurs due to the lack of protective superoxide dismutase that removes superoxide free radicals produced by reactions involving molecular oxygen [29]. Methanogens are abundant in habitats where electron acceptors other than CO_2 are limiting. When these limited electron acceptors, like O_2, NO_3^-, Fe^{3+} and SO_4^{2-}, are present, methanogens are outcompeted by the bacteria that utilize them, thus preventing the occurrence of methanogenesis [24].

In many natural environments containing complex organic compounds, where light, sulfate, and nitrate are limited, the methanogens will cooperate with other chemoheterotrophic bacteria in the process of degrading organic substances. The last step in a series of reactions by which organic compounds are degraded is marked by the production of methane and CO_2; methanogens occupy this terminal position in the breakdown of organic matter. Methanogens can use H_2 and CO_2, as well as formate or acetate, which are produced by fermentative bacteria. Methanogens have also been known to form obligate associations with obligate hydrogen producing acetogenic bacteria; this association is known as syntrophy, taking part in interspecies hydrogen transfer [34]. Eleven species of syntrophic bacteria have been previously described as partnerships of methanogens [29].

In their natural environments, methanogens must compete for substrates that are required and are necessary for methanogenesis. This is especially evident in sulfate and nitrate rich environments. In these environments, the methanogens are outcompeted by sulfate reducing bacteria and nitrate reducing bacteria [35]. These bacteria are more efficient thermodynamically and therefore have a higher affinity for H_2 and higher growth yields [34]. Although methanogens are outcompeted in these environments, small amounts of methane can still be detected. This is most likely because methanogens can utilize substrates that bacteria, like sulfate and nitrate reducing bacteria, cannot include methanol, methylamines, and methionine [35]. By forming symbiotic methanogenic associations and through anaerobic microzones, methanogens can potentially overcome competition for available substrates [28].

Methanogen distribution in natural environments is completely dependent on their ability to adapt to certain factors, like temperature, chemical concentrations, and even flooding regimes. For example, two thirds of biogenic methane are derived from acetate and in environments where there are higher concentrations of acetate, species with higher acetate thresholds tend to prevail [36]. Two genera that can utilize acetate for methanogenesis are the *Methanosaeta* and the *Methanosarcina*. The *Methanosarcina* are obligate acetotrophs, while the *Methanosaeta* are not. They can use acetate, H_2 and CO_2, and formate for methanogenesis. In 1998, Grobkopf and colleagues conducted a study that found that the *Methanosarcina* had a higher threshold for acetate than the *Methanosaeta* [37]. This indicates that the *Methanosarcina* require a higher concentration of acetate in order for methanogenesis to occur. The *Methanosaeta* do not require as high a concentration. *Methanosaeta* use the acetyl CoA synthetase enzyme to activate acetate. This enzyme is functional and effective when there are low acetate concentrations, which is potentially why *Methanosaeta* have a lower acetate threshold. *Methanosarcina*, on the other hand, use acetate kinase to activate acetate and this enzyme is not effective when there are low concentrations of acetate [38]. Other studies conducted have found that acetate concentrations increased with soil depth but methane emissions decrease with soil depth. As soil depth increases, all other available substrates for methanogenesis decrease until acetate is the only substrate left, and there are only certain methanogens that can utilize acetate for methanogenesis. This decreased the amount of methanogenesis occurring thus decreases methane emissions [38].

Another factor that can determine the diversity of methanogens is temperature. Temperature is an important regulator of microbial activity involved in methane production. One study conducted by Chin, *et al*. found that as the temperature decreased from 30°C to 15°C, the amount of methane and H_2 produced also decreased and there was an increase in acetate [39]. When there were decreases in temperature the degradation pathway of organic matter changed and led to an accumulation of acetate, propionate, caproate, lactate and isopropanol. This change also led to a decrease in H_2 partial pressures. This suggested that at lower temperatures acetotrophic methanogenesis predominates. During the initial sampling for this study, they found members of the families *Methanosarcinaceae, Methanobacteriaceae, Methanosaetaceae*, and Euryarchaeotal Rice Clusters I and IV. When these samples were incubated at 15°C they found that same organisms were present but noticed there was an increase in the abundance of *Methanosarcinaceae* and Methanocellales. When the samples were incubated at 30°C the only organisms that were dominant were ones belonging to the family *Methanosarcinaceae* and Euryarchaeotal Rice Cluster I. This illustrates that with the change in temperature there is also a change in the abundance and diversity of methanogenic species [39].

Flood patterns can also have an effect on the microbial community diversity. A study conducted by Kemnitz, *et al*. found that the diversity of methanogenic species increased as flood frequency increased [38]. In sites that were permanently flooded they found members of *Methanomicrobiaceae, Methanobacteriaceae, Methanosarcinaceae, Methanosaetaceae*, and Euryarchaeotal Rice Clluster III, IV, and VI. In sites that were rarely flooded they found only members of

Methanomicrobiaceae and Euryarchaeotal Rice Cluster IV. In this study, they amplified the *mcrA* gene which codes for methyl coezyme-M reductase and can be used as an alternative to 16S rRNA sequences [40]. Kemnitz *et al*. found that amplification of this gene was difficult from sites that were rarely flooded and that almost no PCR product was obtained [38]. This could have been because methanogens were less abundant in these sites. As the flooding frequency increased so did the diversity of Archaea and the abundance of Archaea in that environment.

Studies have also shown that the change in seasons can affect the diversity of species in sediment. Changes have been noted between summer and winter. During summer months, the methanogenic community in the sediment was composed primarily of members from the *Methanosarcinaceae* family. During the winter months, the community was composed of members of the *Methanococcaceae, Methanobacteriaceae* and *Methanothermaceae* families [41].

Methanogens can inhabit a wide variety of environments, including freshwater and marine sediments, marshes, swamps, sludge, rice soils, and intestinal tracts of almost all living creatures. Some of the factors that determine where methanogens will inhabit are known, like pH and temperature, but there are many factors that are likely not yet known. As more research is done regarding Archaeal diversity in the environment, more species will be identified and with that identification there is more we will be able to learn about these unique organisms and the factors that determine their optimal natural environments.

Methanogen community in Kentucky Lake

In 2007, Margaret Grosser characterized the Archaeal community of the littoral zone of the Ledbetter Creek embayment of Kentucky Lake to determine if changes in the community occurred seasonally due to the manipulation of the water level [42]. Sediment samples were taken from five sites within the flood plain of the embayment, one terrestrial site above the littoral zone and one site within the embayment monthly. DNA was extracted and the rRNA genes were amplified using Archaeal 16S rRNA specific primers. The amplified fragments were cloned to separate individuals and well isolated colonies picked and preserved. Plasmid DNA with inserts were isolated and the Archaeal DNA reamplified by PCR. This DNA was subjected to fingerprinting by digestion it with restriction endonucleases and run on polyacrylamide gradient gels. Clones giving the same fingerprints were considered to be the same species. One insert from each fingerprint group was sequenced and the sequences compared to the database of 16S rRNA genes to identify the organism by BLAST search. Eighty-four sequences total were identified. These sequences showed an identity greater than 91% to 10 methanogenic species of Archaea and five species of uncultured Archaea. All the organisms that Grosser identified were methanogens [42]. Grosser also performed community fingerprinting of Archaeal 16S rRNA genes by digesting the amplified sediment DNA from each of her seven sites (five littoral zone sites, one embayment site, and one terrestrial site) monthly for a year and comparing them. These comparisons showed that there were no differences within the littoral zone community (site-to-site) or between the embayment community and the littoral zone community, but there was a significant difference between the terrestrial community and the sediment community.

Grosser also found that there were significant differences in the community between different months throughout the year [42].

In 2011, Xuelian Chen set out to analyze the sequences of the remaining clones that Grosser did not examine to determine whether clones with the same fingerprint were actually the same species [43]. Chen purified and sequenced DNA from 59 clones that were identified as *Methanosaeta concilii*-like. The sequences were then compared to known sequences in the Genebank database by conducting BLAST searches. Chen found that the 59 clones were amplified from 50 different species that fell into the genus Methanosaeta and four other genera [43]. She used a similarity index of 98.7 as the limit for species level and 87 for a limit at the genus level [43].

The purpose of the study was to determine if the Archaeal community of the littoral zone of the Ledbetter Creek embayment at Kentucky Lake was still primarily dominated by the *Methanosaeta concilii*-like species as determined to be in previous studies 9 years earlier. Using the same technique of amplifying rRNA genes, cloning, sequencing, and BLAST analysis the Archaeal species present now will be identified.

Chapter II

Materials and Methods

Throughout the project the water that was used was either included in kits or purchased from Fisher and was molecular biology grade. Other chemicals, not included in kits, were of highest grade available from Fisher or Sigma-Aldrich.

Sample Collection

Sediment samples were collected from the Ledbetter Creek Embayment study site twice a year (Fig. 4). The sample site was located along a transect within the mudflat of the littoral zone. There are five sites total along the transect that are marked by wells that had been placed there for previous studies (Fig. 4). Sediment samples were collected from within two feet of the well that marked the second sample site along the transect. One sample was taken during the summer pool in August 2011 and one sample was taken during the winter pool in January 2012. The top two centimeters were removed, and the samples were taken from the next five centimeters. A sterile 60 ml centrifuge tube was filled with the sediment and the sample was kept on ice and returned to the laboratory where it was kept frozen at -20°C until use.

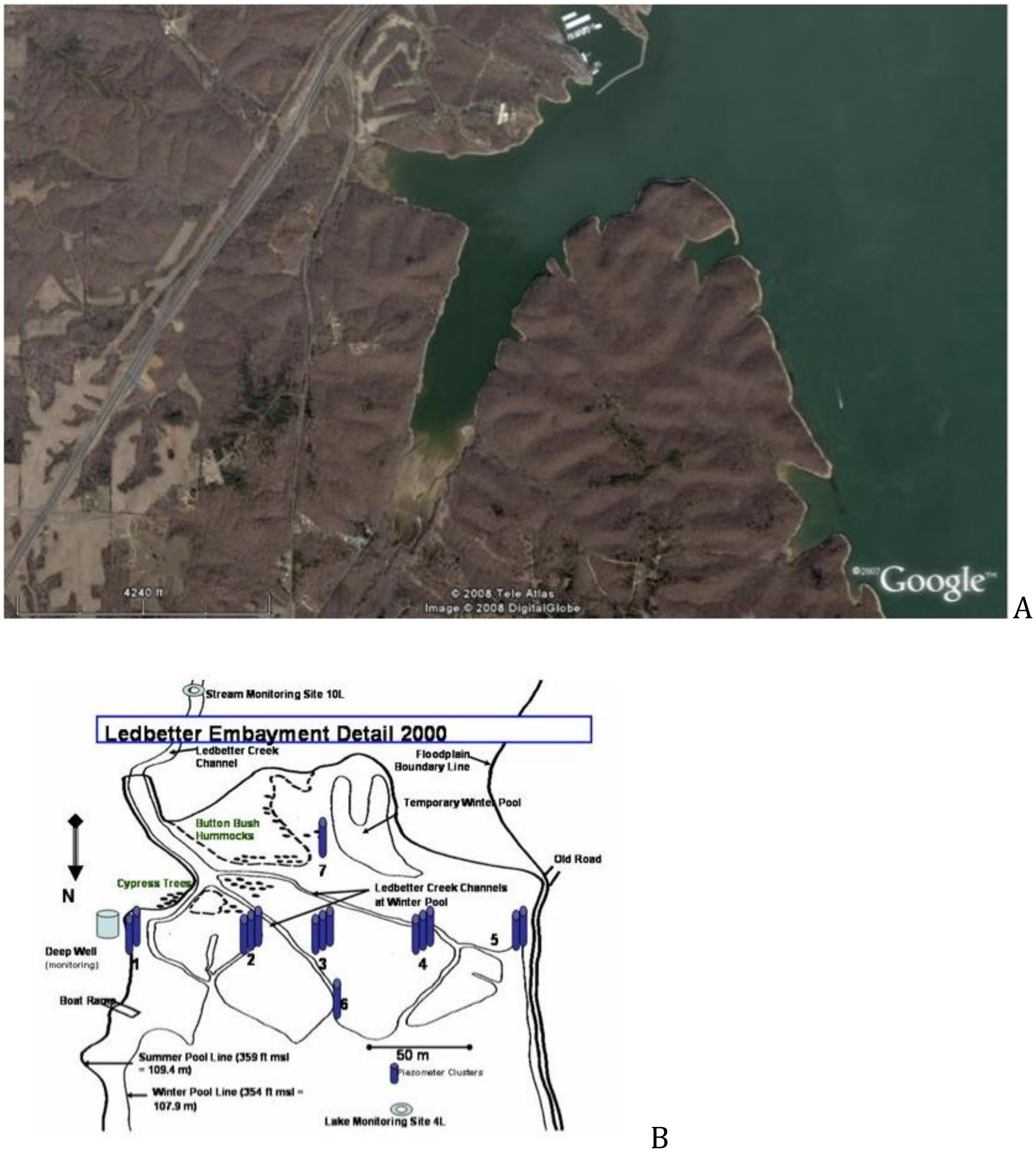


Figure 4: A. Aerial view of the Ledbetter Creek Embayment (Google maps). B. Ledbtter Creek Embayment in more detail. C. shows the locations of the 5 wells that span the littoral zone of the embayment (1). Samples were taken near well-set 2.

DNA Extraction

DNA was extracted from the sediment using a PowerSoil™ DNA Isolation Kit (MoBio) and a BIO 101 Fast Prep Machine set at a speed of 6.0 for 40 seconds. Isolated DNA was quantified, and the resulting quantification ranged from 67 to 112 ng/μl.

PCR Amplification

The extracted DNA was used as the template in Polymerase Chain Reactions (PCR). The amplification was accomplished using nested primers. For the first reaction a 16S rRNA PCR amplification was performed using Archaeal specific primers 1AF and 1404AR (Table 2) [44]. These primers amplify a 1404 base pair fragment of Archaeal specific rRNA genes. The PCR was done using a PCR Optimization Kit (Epicentre). The amplification mixture consisted of 2 μl template (which amounted to roughly 200 ng), 2.5 μL of the 1AF primer and 1404AR primer (both diluted to 20 μM), 2 μl Tfl DNA polymerase (1 unit/μl) (Epicentre), and 16 μl sterile H_2O. This amplification mixture was duplicated 12 times. To each mixture 25 μl of one of the small optimization kit pre-mixes was added giving a total volume of 50 μl. All 12 of the pre-mixes were used (A to L). The reactions were run in a PTC-200 Thermal Cycler (MJ Research). The reaction conditions were as follows: initial denaturation step for five minutes at 94°C followed by touchdown protocol for 20 cycles at 71°C to 61°C with the annealing temperature reduced by 1°C every two cycles, followed by 15 additional cycles at an annealing temperature of 61°c. Denaturation and annealing steps were one minute long. All the extension steps

Table 2: Archaeal specific 16S rRNA primers used in this study [44].

Primer	**Sequence**
1AF	5’ MTCYGKTTGATCCYGSCGRAG 3’
1404AR	5’ CGGTGTGTGCAAGGRGC 3’
100AR	5’ TGGGTCTCGCTCGTTG 3’

were three minutes long with the exception of the final extension cycle, which was ten minutes long [44]. The PCR products were run on a 0.8% agarose gel to ensure the success of the PCR. Amplified samples were purified using the UltraClean PCR CleanUp Kit from MoBio. The samples were again quantified, the results ranging from 86-to 128 ng/µl.

The amplified samples from the first PCR were used as the template for the second PCR. For the second PCR, 16S rDNA PCR amplification was performed using Archaeal specific primers 1AR and 1100AR (Table 1). The primers amplify an 1100 base pair fragment of Archaeal specific rRNA genes. The second PCR also utilized the PCR Optimization Kit (Epicentre). The amplification mixture consisted of 2 µl template, 2.5 µl of primer 1AF and 1100AR (both diluted to 20 µM), 2 µl Tfl DNA Polymerase (Epicentre), and 16 µl reverse osmosis H_2O. This amplification mixture was replicated 12 times. To each mixture 25 µl of one of the Optimization kit pre-mixes was added giving a total volume of 50 µl. All 12 of the pre-mixes were used (A to L). The reactions were run in a PTC-200 Thermal Cycler (MJ Research). The same reaction conditions from the first PCR apply to the second PCR. The PCR products were quantified yielding results that ranged from 421 to 588 ng/µL. The amplified samples were again run on a 0.8% agarose gel to confirm the success of the PCR.

Cloning

The products from the second PCR were ligated into the pGEM®-T Easy Vector (Promega). The ligation mixture was as follows: 1 µl PCR product, 1 µl pGEM®-T Easy Vector (50 ng/µl), 1 µl T4 DNA Ligase (3 units/µl), 5 µl 2X Ligation

Buffer, and 2 μl reverse osmosis H_2O to give a total reaction volume of 10 μl. The ligation mixture was then used to transform competent DH5α *E. coli* cells (New England Biolabs). Transformants were plated on Luria broth (LB)-Amp plates (25μg/ml of Amp) that contained Xgal (20 mg/ml) and IPTG (23.8 mg/ml), and blue/white colony screenings were performed.

White colonies growing on the LB-Amp, Xgal plates were used to make up overnight cultures. Plasmid DNA was isolated from overnight cultures using an UltraClean 6 Minute Mini Prep Kit (MoBio). Resulting samples were quantified yielding results that ranged from 50 to 500 ng/μl. Stocks were also made using the overnights by adding 1.0ml of overnight to 0.5ml of a 50% glycerol solution.

Five hundred ng of the purified plasmid DNA products were digested using EcoR1 (20,000 units/ml) (New England BioLabs®) restriction enzyme at 37°C for one hour. The digested samples were run on a 0.8% agarose gel to confirm size of insert.

DNA Sequencing

Inserts from plasmid that contained inserts measuring 1100 base pairs were sequenced. A total of 50 summer clones and 50 winter clones were sequenced. The summer and winter clones were sent to SeqWright DNA Technology Services in Houston, Texas to be sequenced. Although 100 total clones were sequenced, usable data for only 43 summer clones and 20 winter clones were obtained.

DNA inserts were sequenced from both ends, merged and edited using MacVector Software. Edited sequences were then used in NCBI database BLAST searches to attempt to identify the organism. Similarity matrices were constructed to determine how the sequences related to each other and how they related to sequences identified by Maggie Grosser and those sequenced by Xuelian Chen. Similarity matrices were conducted using MacVector and Geneious Software. A phylogenetic tree was constructed to determine also how these new sequences related to each other using MacVector Software.

Chapter III

Results

ANALYSIS OF INDIVIDUAL ARCHAEA

Sequencing Analysis

One hundred clones were isolated in total. SeqWright Incorporated sequenced all 100 clones. Seven of the summer clones and 30 of the winter clones could not be sequenced and were thrown out, leaving 63 total sequences. Merged sequences were edited and aligned using MacVector (MacVector Inc, North Carolina, USA) and Geneious software (Biomatters Limited, New Zealand). Edited and aligned DNA sequences were compared with known sequences in the NCBI database through BLAST searches. The BLAST results for summer and winter clones are summarized in tables 3 and 4. Clone sequences were matched to known organisms based on similarity index. For alignment and BLAST searches gap penalties of 5 and 2 were chosen based on previous studies [45, 46].

Based on the BLAST data, the summer and winter clones were split into eight different groups based on which organisms their sequences matched in the database, these data are listed in table 5. Twenty-two of the 63 sequences, 34%, belong to the genus *Methanosaeta.* Seventeen clones, 26.9%, belong to the genus *Methanoregula*. One clone, 1.6%, belonged to the *Methanospirillum* genus. Eighteen clones, 28.6%, belong to the genus *Methanosphaerula*.

Table 3: BLAST search results for the summer clones. Compares clones isolated to known organism sequences in the NCBI database.

Clone	BLAST Match	Percent Identity	Number of nucleotides matched	Number of Gaps	Accession number
S1-2011	*Methanosphaerula palustris*	93	849/917	0	NC 011832.1
S2-2011	*Methanosphaerula palustris*	92	954/1038	2	NC 011832.1
S3-2011	*Methanosaeta concilii*	97	1012/1038	0	NC 015416.1
S4-2011	*Methanosaeta concilii*	98	1018/1044	0	NC 015416.1
S5-2011	*Methanosaeta concilii*	97	1013/1041	0	NC 015416.1
S6-2011	*Methanosaeta concilii*	97	964/992	0	NC 015416.1
S7-2011	*Methanosphaerula palustris*	94	972/1037	0	NC 011832.1
S8-2011	*Methanosphaerula palustris*	95	981/1036	0	NC 011832.1
S9-2011	*Methanosphaerula palustris*	92	921/1001	2	NC 011832.1
S10-2011	*Methanoregula boonei*	95	986/1039	2	NC 009712.1
S11-2011	*Methanoregula boonei*	96	996/1038	0	NC 009712.1
S12-2011	*Methanosphaerula palustris*	92	948/1033	2	NC 011832.1
S13-2011	*Methanosphaerula palustris*	92	950/1035	2	NC 011832.1
S14-2011	*Methanosphaerula palustris*	92	955/1038	1	NC 011832.1
S15-2011	*Methanosphaerula palustris*	92	956/1039	1	NC 011832.1
S16-2011	*Methanosaeta concilii*	98	1018/1041	0	NC 015416.1
S17-2011	*Methanosaeta concilii*	98	1018/1041	0	NC 015416.1
S18-2011	*Methanoregula boonei*	95	979/1038	0	NC 009712.1
S19-2011	*Methanoregula boonei*	94	979/1038	0	NC 009712.1
S20-2011	*Methanoregula boonei*	97	1003/1038	0	NC 009712.1
S21-2011	*Methanoregula boonei*	95	983/1032	0	NC 009712.1
S22-2011	*Methanoregula boonei*	95	983/1032	0	NC 009712.1
S23-2011	*Methanosaeta concilii*	98	1021/1039	0	NC 015416.1

S24-2011	*Methanosphaerula palustris*	92	954/1039	1	NC 011832.1
S26-2011	*Methanosaeta concilii*	98	814/832	3	NC 015416.1
S27-2011	*Methanosphaerula palustris*	88	572/653	6	NC 011832.1
S28-2011	*Methanosphaerula palustris*	91	829/906	7	NC 011832.1
S29-2011	*Methanosaeta concilii*	98	1023/1049	4	NC 015416.1
S31-2011	*Methanosphaerula palustris*	92	883/962	4	NC 011832.1
S32-2011	*Methanosphaerula palustris*	92	869/946	4	NC 011832.1
S33-2011	*Methanocella paludicola*	92	845/919	7	NC 013665.1
S34-2011	*Methanosphaerula palustris*	93	973/1047	4	NC 011832.1
S35-2011	*Methanoregula boonei*	95	582/615	2	NC 009712.1
S36-2011	*Methanoregula boonei*	94	875/931	3	NC 009712.1
S39-2011	*Methanosphaerula palustris*	91	631/696	7	NC 011832.1
S40-2011	*Methanosaeta concilii*	90	828/924	6	NC 015416.1
S41-2011	*Methanoregula boonei*	93	775/837	3	NC 009712.1
S42-2011	*Methanosaeta concilii*	95	553/580	1	NC 015416.1
S43-2011	*Methanosaeta concilii*	94	775/841	1	NC 015416.1
S44-2011	*Methanoregula boonei*	94	896/956	4	NC 009712.1
S45-2011	*Methanosphaerula palustris*	91	738/815	3	NC 011832.1
S47-2011	*Uncultured methanogenic archaeon RC-I*	95	856/917	2	NC 009464.1
S49-2011	*Methanoregula boonei*	95	937/1028	5	NC 009712.1

Table 4: BLAST search results for winter clones. Compares the winter clone sequences with known organism sequences in NCBI database.

Clone	BLAST Match	Percent Identity	Number of nucleotides matched	Number of Gaps	Accession number
W8-2012	*Methanosaeta concilii*	89	645/727	8	NC 015416.1
W10-2012	*Methanosaeta concilii*	95	1000/1050	3	NC 015416.1
W12-2012	*Methanoregula boonei*	94	519/552	2	NC 009712.1
W15-2012	*Methanoregula boonei*	93	895/964	6	NC 009712.1
W22-2012	*Methanosaeta harundinacea*	84	697/825	8	NC 017527.1
W24-2012	*Methanosaeta concilii*	97	1020/1047	0	NC 015416.1
W25-2012	*Methanosaeta concilii*	95	998/1046	3	NC 015416.1
W28-2012	*Methanoregula boonei*	94	766/818	4	NC 009712.1
W29-2012	*Methanosaeta concilii*	94	900/956	4	NC 015416.1
W31-2012	*Methanosaeta concilii*	94	876/935	6	NC 015416.1
W32-2012	*Methanosaeta concilii*	97	1019/1050	3	NC 015416.1
W34-2012	*Methanocaldococcus vulcanis*	78	543/700	6	NC 013407.1
W35-2012	*Methanoregula boonei*	90	889/983	8	NC 009712.1
W36-2012	*Methanosaeta concilii*	94	839/888	4	NC 015416.1
W37-2012	*Methanohalophilus mahii*	86	818/952	5	NC 014002.1
W38-2012	*Methanosphaerula palustris*	92	733/800	6	NC 011832.1
W40-2012	Uncultured methanogenic archaeon RC-I	94	755/805	5	NC 009464.1
W41-2012	*Methanospirillum hungatei*	95	740/781	0	NC 007796.1
W44-2012	*Methanoregula boonei*	94	869/923	6	NC 009712.1
W50-2012	*Methanosaeta concilii*	94	850/904	6	NC 015416.1

Table 5: Grouping of clones and most closely matched organisms based on genus.

Group	**Genus matched**	**Number of clones matched**
1	*Methanoregula*	17
2	*Methanospirillum*	1
3	*Methanosphaerula*	18
4	*Methanosaeta*	22
5	Uncultured methanogenic archaeon RC-1	2
6	*Methanocella*	1
7	*Methanohalophilus*	1
8	*Methanocaldococcus*	1

One clone was closely related to the genus *Methanohalophilus* with 86% identity.

The closest match for one clone was the genus *Methanocaldococcus* but with only

78% identity. One clone belonged to the genus *Methanocella.* The final two clones shared 94% and 95% identity with uncultured methanogenic archaeon RC-I. Sequence percent identities ranged from 78 to 98. For two sequences to be considered the same species, they should have a percent identity that is greater than or equal to 98.7 [47]. There were 6 that were close, having a percent identity of 98. For sequences to be assigned to the same genera, they must have a sequence percent identity of 87 or higher [47]. Sixty of the clones had sequence percent identities high enough, 87% or greater, to match them with known genera. Three of the clones had sequence similarity indices of less than 87 and could represent novel genera.

Sequence Similarities Between Current and Previous Findings

Similarity matrices were set up to compare the sequences against each other to determine how similar the clone sequences were. Summer and winter clones were compared first. Table 6 is a summary of the similarity matrix for each summer and winter clone. Sequences compared with itself had a percent identity of 100%. There was a total of 31 clones that had a percent identity of 87% or greater and there were only 6 clones that had a percent identity of 98.7% or greater.

Clones that belong with the genus *Methanosaeta* were compared to clones sequenced by Margaret Grosser in 2007 and Xuelian Chen in 2011, primarily because both showed *Methanosaeta concilii*-like organisms were the most abundant.

Table 6: Sequence similarity matrix summary for summer and winter clones. The full table is in the Appendix (table A1).

	Number of clones similar/total clones
Clone sequences that had percent identities of 87% or greater to other summer and winter clones	31/63
Clone sequences that had percent identities of 98.7% or greater to other summer and winter clones	6/63

Table 7 is the summary for the similarity matrix that compares clones that most closely related *Methanosaeta concilii* and the clones isolated by Grosser were *Methanosaeta concilii*-like. In total, 9 of the 22 summer and winter clones that belong to the genus *Methanosaeta* showed 87% similarity with the Grosser clones and zero showed similarity of 98.7%.

Most of the clones sequenced by Chen in 2011 belonged to the genus *Methanosaeta*. All of Chen's clones were compared to summer and winter clones that most closely matched *Methanosaeta* from this study. Table 8 is a summary of the similarity matrix that compared summer and winter clones that closely matched *Methanosaeta concilii* and clones sequenced by Chen that also closely matched *Methanosaeta concilii*. Thirteen of the 22 summer and winter clones compared to Chens clones showed a similarity of 87% while zero showed similarity of 98.7%.

The clones sequenced by Grosser matched more organisms than just *Methanosaeta concilii*. Clones that matched organisms other than *M. concilii* were compared with the different clone groups that were isolated during summer and winter pools. When Grosser's study was done, *Methanoregula boonei* was not in the NCBI database, but some of her clones closely matched *Methanospirillum* species. Summer and winter clones that matched the *Methanoregula* genus also has a high percent identity to *Methanospirillum* and *Methanosphaerula* genera, but it was less than 87%. The clones that Grosser sequenced that most closely matched *Methanospirillum* species were then compared to clones in groups 1-3. The summary for these comparisons is listed in table 9.

Table 7: Sequence similarity matrix summary for summer and winter clones that closely matched *Methanosaeta concilii* and compared with clones isolated by Margaret Grosser that also closely matched *Methanosaeta concilii*. The full table is located in the Appendix (table A2).

	Number of similarities
Summer and winter clone sequences with a similarity index of 87 or greater when compared to Grosser's clone sequences	9
Summer and winter clone sequences with a similarity index of 98.7 or greater when compared to Grosser's clone sequences	0

Table 8: Sequence similarity matrix summary for summer and winter clones that closely matched *Methanosaeta concilii* and compared with clones isolated by Xuelian Chen that also closely matched *Methanosaeta concilii*. The full table is located in the Appendix (table A3).

	Number of Sequence Combinations
Summer and winter clone sequences with a similarity index of 87 or greater when compared to Chens's clone sequences	13
Summer and winter clone sequences with a similarity index of 98.7 or greater when compared to Chens's clone sequences	0

Out of the 36 summer and winter clones that groups 1-3 were comprised of, only 17 showed sequence similarity of 87% to clones from Grossers study that belong to the genus *Methanospirillum* and zero clones had a similarity of 98.7% to the Grosser clones.

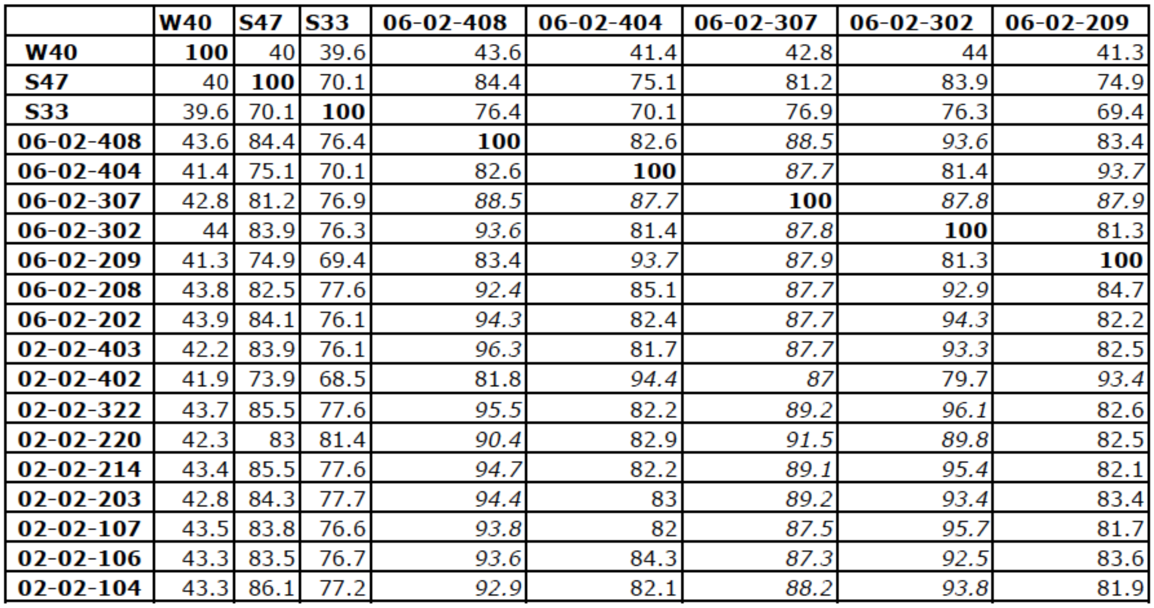
Summer and winter clones that had percent identity of 87% or more with uncultured methanogenic archaeon or the genus *Methanocella* were compared with clones sequenced by Grosser that matched uncultured methanogenic archaeon. Table 10 shows the comparison results for these sequences. Of the 3 summer and winter clones that were compared to clones isolated by Grosser, zero had a similarity index of 87% or more.

The last comparison was between summer and winter clones that had a percent identity of 87% with the genus *Methanohalophilus* and Grosser's clones that belonged to the same genus. Table 11 lists the results of this comparison. There were only 2 clones from the current study that were part of this comparison and neither of them showed enough similarity with Grossers clones to be considered the same organisms.

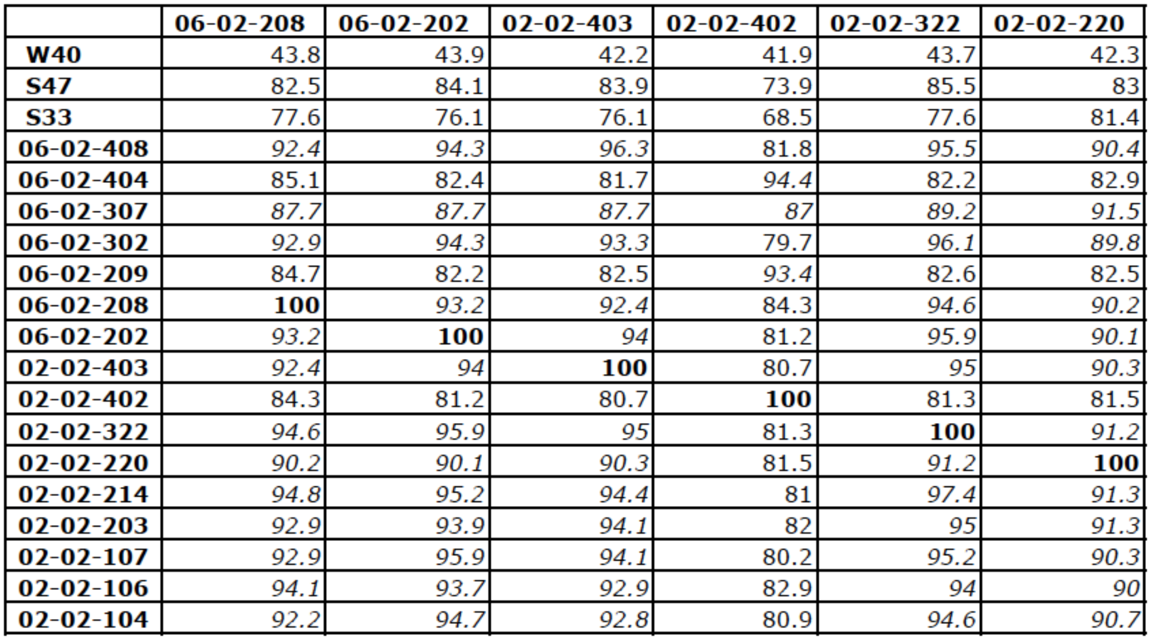
Table 9: Sequence similarity matrix summary for group 1 clones compared with clones isolated by Margaret Grosser that closely matched *Methanospirillum*. The full table is in the Appendix (table A4).

	Number of clones similar
Group 1 clone sequences with a similarity index greater than 87 when compared to Grossers's clone sequences	17
Group 1 clone sequences with a similarity index greater than 98.7 when compared to Grossers's clone sequences	0

Table 10: Comparison of summer and winter clones that closely matched Unculutured methanogenic archaeon with Grosser's clones that also closely matched uncultured methanogenic archaeon. Italicized numbers indicate a similarity index of 87 or greater; bolded numbers indicate sequences with a similarity index of 98.7 or greater.



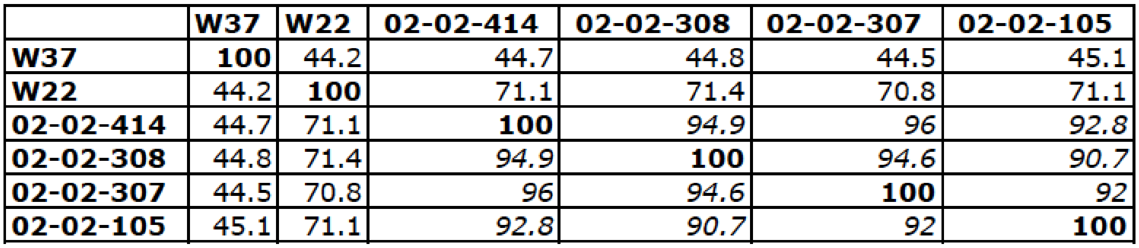
	W40	**S47**	**S33**	**06-02-408**	**06-02-404**	**06-02-307**	**06-02-302**	**06-02-209**
W40	**100**	40	39.6	43.6	41.4	42.8	44	41.3
S47	40	**100**	70.1	84.4	75.1	81.2	83.9	74.9
S33	39.6	70.1	**100**	76.4	70.1	76.9	76.3	69.4
06-02-408	43.6	84.4	76.4	**100**	82.6	*88.5*	*93.6*	83.4
06-02-404	41.4	75.1	70.1	82.6	**100**	*87.7*	81.4	*93.7*
06-02-307	42.8	81.2	76.9	*88.5*	*87.7*	**100**	*87.8*	*87.9*
06-02-302	44	83.9	76.3	*93.6*	81.4	*87.8*	**100**	81.3
06-02-209	41.3	74.9	69.4	83.4	*93.7*	*87.9*	81.3	**100**
06-02-208	43.8	82.5	77.6	*92.4*	85.1	*87.7*	*92.9*	84.7
06-02-202	43.9	84.1	76.1	*94.3*	82.4	*87.7*	*94.3*	82.2
02-02-403	42.2	83.9	76.1	*96.3*	81.7	*87.7*	*93.3*	82.5
02-02-402	41.9	73.9	68.5	81.8	*94.4*	*87*	79.7	*93.4*
02-02-322	43.7	85.5	77.6	*95.5*	82.2	*89.2*	*96.1*	82.6
02-02-220	42.3	83	81.4	*90.4*	82.9	*91.5*	*89.8*	82.5
02-02-214	43.4	85.5	77.6	*94.7*	82.2	*89.1*	*95.4*	82.1
02-02-203	42.8	84.3	77.7	*94.4*	83	*89.2*	*93.4*	83.4
02-02-107	43.5	83.8	76.6	*93.8*	82	*87.5*	*95.7*	81.7
02-02-106	43.3	83.5	76.7	*93.6*	84.3	*87.3*	*92.5*	83.6
02-02-104	43.3	86.1	77.2	*92.9*	82.1	*88.2*	*93.8*	81.9



	06-02-208	**06-02-202**	**02-02-403**	**02-02-402**	**02-02-322**	**02-02-220**
W40	43.8	43.9	42.2	41.9	43.7	42.3
S47	82.5	84.1	83.9	73.9	85.5	83
S33	77.6	76.1	76.1	68.5	77.6	81.4
06-02-408	*92.4*	*94.3*	*96.3*	81.8	*95.5*	*90.4*
06-02-404	85.1	82.4	81.7	*94.4*	82.2	82.9
06-02-307	*87.7*	*87.7*	*87.7*	*87*	*89.2*	*91.5*
06-02-302	*92.9*	*94.3*	*93.3*	79.7	*96.1*	*89.8*
06-02-209	84.7	82.2	82.5	*93.4*	82.6	82.5
06-02-208	**100**	*93.2*	*92.4*	84.3	*94.6*	*90.2*
06-02-202	*93.2*	**100**	*94*	81.2	*95.9*	*90.1*
02-02-403	*92.4*	*94*	**100**	80.7	*95*	*90.3*
02-02-402	84.3	81.2	80.7	**100**	81.3	81.5
02-02-322	*94.6*	*95.9*	*95*	81.3	**100**	*91.2*
02-02-220	*90.2*	*90.1*	*90.3*	81.5	*91.2*	**100**
02-02-214	*94.8*	*95.2*	*94.4*	81	*97.4*	*91.3*
02-02-203	*92.9*	*93.9*	*94.1*	82	*95*	*91.3*
02-02-107	*92.9*	*95.9*	*94.1*	80.2	*95.2*	*90.3*
02-02-106	*94.1*	*93.7*	*92.9*	82.9	*94*	*90*
02-02-104	*92.2*	*94.7*	*92.8*	80.9	*94.6*	*90.7*

	02-02-214	**02-02-203**	**02-02-107**	**02-02-106**	**02-02-104**
W40	43.4	42.8	43.5	43.3	43.3
S47	85.5	84.3	83.8	83.5	86.1
S33	77.6	77.7	76.6	76.7	77.2
06-02-408	*94.7*	*94.4*	*93.8*	*93.6*	*92.9*
06-02-404	82.2	83	82	84.3	82.1
06-02-307	*89.1*	*89.2*	*87.5*	*87.3*	*88.2*
06-02-302	*95.4*	*93.4*	*95.7*	*92.5*	*93.8*
06-02-209	82.1	83.4	81.7	83.6	81.9
06-02-208	*94.8*	*92.9*	*92.9*	*94.1*	*92.2*
06-02-202	*95.2*	*93.9*	*95.9*	*93.7*	*94.7*
02-02-403	*94.4*	*94.1*	*94.1*	*92.9*	*92.8*
02-02-402	81	82	80.2	82.9	80.9
02-02-322	*97.4*	*95*	*95.2*	*94*	*94.6*
02-02-220	*91.3*	*91.3*	*90.3*	*90*	*90.7*
02-02-214	**100**	*94.8*	*94.9*	*93.8*	*94*
02-02-203	*94.8*	**100**	*93.4*	*93.1*	*93.3*
02-02-107	*94.9*	*93.4*	**100**	*93.9*	*94.1*
02-02-106	*93.8*	*93.1*	*93.9*	**100**	*92.9*
02-02-104	*94*	*93.3*	*94.1*	*92.9*	**100**

Table 11: Comparison of winter clones and Grosser's clones that most closely matched *Methanohalophilus.* Italicized numbers indicate a similarity index of 87 or greater; bolded numbers indicate sequences with a similarity index of 98.7 or greater.



	W37	**W22**	**02-02-414**	**02-02-308**	**02-02-307**	**02-02-105**
W37	**100**	44.2	44.7	44.8	44.5	45.1
W22	44.2	**100**	71.1	71.4	70.8	71.1
02-02-414	44.7	71.1	**100**	*94.9*	*96*	*92.8*
02-02-308	44.8	71.4	*94.9*	**100**	*94.6*	*90.7*
02-02-307	44.5	70.8	*96*	*94.6*	**100**	*92*
02-02-105	45.1	71.1	*92.8*	*90.7*	*92*	**100**

Phylogenetic Analysis

A phylogenetic tree was constructed to determine the phylogenetic relationship between summer and winter clones. The tree was constructed using MacVector software. Figure 5 shows the complete phylogenetic tree. There were three large clusters shown on the tree. The first cluster, A, are most closely related *Methanosaeta concilii* in the NCBI BLAST searches. This cluster also contained the clones that most closely matched the uncultured methanogenic archaeon. Cluster B were most closely related to *Methanosphaerula palustris*. Cluster C were closely related *Methanoregula boonei* and *Methanosphaerula palustris*. There were also clones in this cluster that were similar to *Methanosaeta concilii* and one clone in this cluster was similar to an uncultured methanogenic archaeon as determined by the BLAST searches. There were also a few small clusters, which mainly consisted of pairs of clones that were closely related. The first small cluster pair, D, was S19 and S18, which was most closely related to *Methanoregula boonei*. The second cluster pair, E, consisted of S7 and S8 and these sequences were most closely related to *Methanosphaerula palustris*. The third small cluster, F, consisted of S10, S11 and S21 and this cluster was most closely related to *Methanoregula boonei*. The fourth small cluster pair, G, consisted of W15 and S41, which were most closely related to *Methanoregula boonei*.

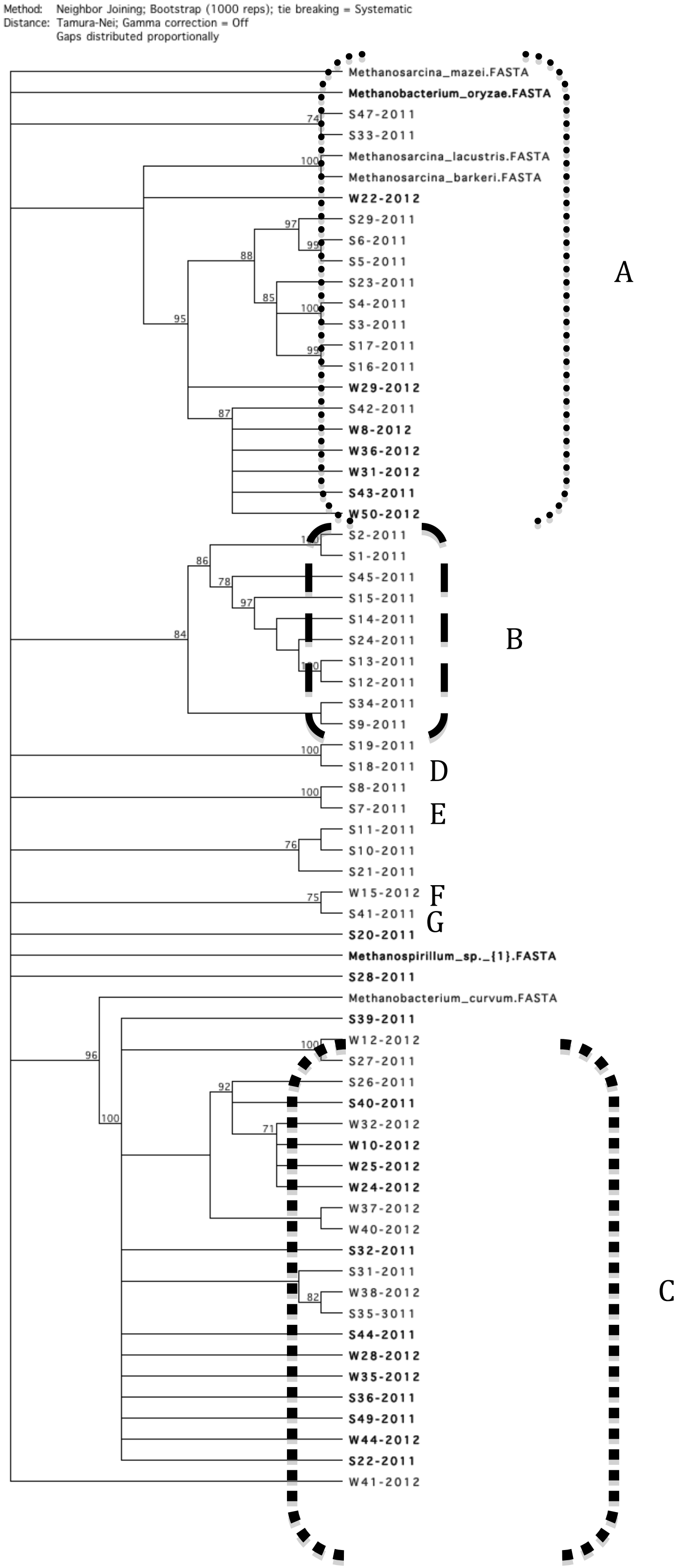

Method: Neighbor Joining; Bootstrap (1000 reps); tie breaking = Systematic
Distance: Tamura-Nei; Gamma correction = Off
Gaps distributed proportionally
Methanosarcina_mazei.FASTA
Methanobacterium_oryzae.FASTA
S47-2011
S33-2011
Methanosarcina_lacustris.FASTA
Methanosarcina_barkeri.FASTA
W22-2012
S29-2011
S6-2011
S5-2011
S23-2011
S4-2011
S3-2011
S17-2011
S16-2011
W29-2012
S42-2011
W8-2012
W36-2012
W31-2012
S43-2011
W50-2012
S2-2011
S1-2011
S45-2011
S15-2011
S14-2011
S24-2011
S13-2011
S12-2011
S34-2011
S9-2011
S19-2011
S18-2011
S8-2011
S7-2011
S11-2011
S10-2011
S21-2011
W15-2012
S41-2011
S20-2011
Methanospirillum_sp._{1}.FASTA
S28-2011
Methanobacterium_curvum.FASTA
S39-2011
W12-2012
S27-2011
S26-2011
S40-2011
W32-2012
W10-2012
W25-2012
W24-2012
W37-2012
W40-2012
S32-2011
S31-2011
W38-2012
S35-3011
S44-2011
W28-2012
W35-2012
S36-2011
S49-2011
W44-2012
S22-2011
W41-2012
74
100
97
99
88
85
100
99
95
87
86
78
97
84
100
100
100
76
75
96
100
92
71
82
A
B
D
E
F
G
C

Figure 5: Phylogenetic tree for summer and winter clones. Three large clusters and four small clusters were observed. Clones that most closely related the same or similar Archaea in the NCBI database tended to cluster together. *Methanobacterium curvum*, Methanospirillum sp., *Methanosarcina lacustris, Methanosarcina barkeri, Methanosaeta concilii* and *Methanosarcina mazei* sequences were used to root the tree.

Chapter IV

Discussion

Soil is considered to be the most microbially diverse environment on earth and until recently, Bacteria were the only prokaryotes that had been studied in soil environments. It has become evident that Archaea are also abundant and ubiquitous in soil [48]. This study examined the Archaeal community in sediments of a reservoir littoral zone compared to what was there seven years earlier. There were nine genus or species that 16S rDNA sequences matched in the NCBI database total. Over half of the summer samples, 67%, closely matched group one, which included *Methanoregula boonei and Methanosphaerula palustris.* Twenty eight percent closely matched group 2, which included *Methanosaeta concilii, Methanosaeta thermophila,* and *Methanosaeta harundinacea.* For the winter samples 35% of the clones closely matched the group one organisms. Fifty percent of the clones closely matched the group two organisms. Overall 57% of the clones isolated closely matched the group one organisms and 35% closely matched the group two organisms. In 2007 it was found that 37 of 57, or 65%, summer clones closely matched *Methanosaeta* sp. and three of 77, or 5%, clones matched *Methanospirillum* species. For winter, Grosser found that 38 of 77, or 49%, clones matched *Methanosaeta* sp. and ten of 77, or 13%, clones matched *Methanospirillum* sp [42]. Overall 56% of Grosser's clones closely matched *Methanosaeta* sp. and ten percent closely matched *Methanospirillum* sp. Currently, instead of only one class of methanogens being dominant in the sediment of the Ledbetter Creek embayment, there are two. *Methanoregula boonei and Methanosphaerula palustris* both belong to the Methanomicobia class and the Methanomicrobiales order of Euryarchaeota. Both are recently described species, *Methanoregula boonei* was described in 2008 and *Methanosphaerula palustris* was described in 2009. Members belonging to this order of Euryarchaeota are capable of producing methane from formate and some secondary alcohols. The *Methanosaeta* sp. belongs to the Methanosarcinales order and Methanosaetace family. These organisms are obligate acetotrophic methanogens. For these organisms, acetate is the sole substrate used in methanogenesis [29].

There were less dominant organisms found that were not included in groups one and two. One winter clone closely matched *Methanohalophilus mahii*, which belongs to the family Methanosarcinaceae. These organisms are acetotrophic methanogens and are the most versatile methanogens, having the ability to utilize acetate, H_2+CO_2 and methyl compounds in methanogenesis [29]. One winter clone closely matched *Methanocaldococcus vulcanis*, which belongs to the family Methanocaldococcaceae. These organisms utilize H_2+CO_2 and methyl compounds for methanogenesis [29].

In comparing the data found in summer and winter, it is apparent that there was a shift in the abundance of methanogenic orders found. During the summer months, the Methanomicrobiales order is the most dominant with 67% of the clones sequenced matching organisms from this order. Only 28% of the summer clones sequenced matched the *Methanosaeta* sp. During the winter the *Methanosaeta* sp. are the more dominant with 50% of sequenced clones matching these species. Only

35% of the winter clones sequenced matched the Methanomicrobiales order. There are two factors that could be responsible for the difference in abundance of organisms between summer and winter. One is the availability of substrates for methanogenesis. The restriction of substrates available can limit the abundance of methanogens found in the littoral zone sediment. Many of the organisms matched can utilize H_2+CO_2 and formate, which could lead to competition between organisms. Competition could limit the abundance of these organisms. Both the *Methanosaeta* sp. and *Methanohalopjilus mahii* can utilize acetate. *Methanosaeta* sp. use acetyl coA synthetase in methanogenesis which can be activated in low concentration of acetate, where the Methanosarcinaceae family use acetate kinase, which required higher concentrations of acetate before it can be activated [38].

The second factor, which plays into the first, is temperature. When the temperature decreases there is an increase in the amount of acetate, propionate, caproate, lactate and isoporponal in an environment [40]. This affects the substrates that are available for methanogenesis [39]. There is an increase in the amount of *Methanosaeta* sp. present between summer and winter, winter having the higher amount. This could be due to the increase in acetate available in the environment. There is a higher amount of species belonging to the Methanomicrobiales order in the summer, which could be due to the limited amount of acetate available as well as a higher number of other substrates available.

When current data was compared with Grosser's data, it is apparent that there is also a shift in the abundance of genus and species of methanogens found in the littoral zone sediment. There are many conditions that could be responsible for the shift in organisms over time which includes, but is not limited to, pH, particle size, organic carbon content, nutrient availability, water content, and oxygen concentration [48]. The number of organisms found that belong to the Methanomicrobiales order is higher than it was seven years ago. Grosser found that only ten percent of her clones matched *Methanospirillum* sp., which also belong to the Methanomicrobiales order. One factor that could be responsible for this increase is an increase in substrates utilized by these organisms for methanogenesis. Another possibility to consider is that Grosser had more clones that matched unknown organism*. Methanoregula boonei* and *Methanosphaerula palustris* were not described until after Grosser's thesis was written. Grosser's unknowns could possibly have been these species. There has also been a decline in the number of *Methanosaeta* sp., which could be due to a limited number of substrates present in the environment.

The phylogenetic tree (Fig. 5) was constructed to look at the relationships between the clones of summer and winter samples. The two clones that matched the uncultured methanogenic archeaon RC-I were present in two different clusters. W40-2012 was in cluster C, which consisted of clones that closely matched the Methanomicobia class and the Methanomicrobiales order of Euryarchaeota, which includes *Methanoregula boonei* and *Methanosphaerula palustris*, and S47-2011 was located in cluster A, which contained clones that matched *Methanosaeta concilii*.

Similarity matrices were also constructed to look at relationships between summer and winter clones as well as to determine relationships between clones sequenced by Maggie Grosser and Xuelian Chen (tables 7-11). For sequences to be considered the same species they need to be have a similarity index of at least 98.7%. It was previously accepted that an index of 97% was sufficient to determine when two sequences were the same species, but in 2006 it was determined that 97% was too low and that by using 97% as the cutoff there would be an increase in error [49]. For organisms to be considered part of the same genera there needs to be a similarity index of 87% or higher. Different similarity matrices were constructed to compare summer and winter clones from this study to each other as well as to compare summer and winter clones with clones sequenced by Grosser and Chen. When summer and winter clones were compared (table 6), there were incidences where clone sequences had similarity index of 87 or greater. Few of the clones isolated in this study showed a similarity index of 87% with clones sequenced by Grosser and Chen and none of the current clones were similar enough to be considered the same species. This indicates that the organisms present in the littoral zone sediment currently are different from those that were present seven years ago. Over the past seven years, the environment at the Ledbetter Creek Embayment has altered in a manner that resulted in two classes of Archaea becoming dominant, whereas previously, only one of the two classes were dominant. Methanomicobia has joined Methanosaetace as the dominant classes of Archaea. Since the 2007 study by Maggie Grosser, *Methanoregula boonei* and *Methanosphaerula palustris* have been described. Copelans *et al*. discovered *Methanoregula boonei* in 2007, and Lucas *et al.* discovered *Methanosphaerula palustris* in 2008 [50]. Both Archaeal species belong to the Methanomicobia class and are the two-dominant species from this class in the littoral zone sediment. In

2007, Grosser sequences 29 unidentified archaeal strains and there is a possibility that some are *Methanoregula boonei* or *Methanosphaerula palustris*, but the prospect that the species would show dominance in the study as it does in the current study is unlikely. Methanogen-producing bacteria are studied in only a handful of areas throughout the world because they are incredibly difficult to culture in a laboratory setting. More studies will need to be done on the effects of methanogens on flora and fauna, as well as specific niches for the bacteria itself before further analysis of these results can be completed. Future studies can look at the long-term dominance patterns of *Methanosaeta concilii*-like, *Methanoregula boonei*-like and *Methanosphaerula palustris*-like organisms in the Ledbetter Creek Embayment along with environmental characteristics. These results could be compared to similar studies done in other areas containing the same class of archaea.

Chapter V

Appendix

Sequences

S1-2011
CCGGGAAACTGGGGATAATACTCCATGGGCTACGGAAGCTGGAAGGCTCTGTAACCCAA
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S2-2011
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S3-2011
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S4-2011
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S5-2011
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S6-2011
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S7-2011
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S8-2011
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S9-2011
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S10-2011
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S11-2011
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S12-2011
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S13-2011
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S14-2011
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S15-2011
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S16-2011
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S17-2011
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S18-2011
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S19-2011
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S20-2011
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S21-2011
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S22-2011
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S23-2011
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S24-2011
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S26-2011
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S27-2011
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S28-2011
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S29-2011

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S31-2011
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S32-2011
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S33-2011
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S34-2011
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S35-2011
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S36-2011
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S38-2011
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S39-2011
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S40-2011
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S41-2011
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S42-2011
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S43-2011
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S44-2011
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S45-2011
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S47-2011
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S49-2011
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W8-2012
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W10-2012
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W12-2012
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W15-2012
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W22-2012
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W24-2012
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W25-2012
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W28-2012
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CGGCGATAAGGGAACCTCGAGTGCCAGGATACAATCCTGGCTGTCGTAATGCCTAAAAAG
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GTGCGACCGTGGTCGGTGCCGTAGGGAAGCCGTGAACCGAGCCACCTGGGAAGTACGGGC
CCCAAGGCTGAAACTTAAAGGAATTGGCGGGGGAGCACTACAACGGGTGGGAGCTTGCG
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W31-2012
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GCCACCTGGGAAGTACGGCCGCAAGGGTTGAACTTTAAAGGAATGGGCGGGGGAACACC
CCAACGGGTGGAGCTTGGGGTTTTATTGGGATTCAACCCCGGAAAACTTTACCGGGACCC
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GCTGGGCCC

W32-2012
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W34-2012
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W35-2012
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W36-2012
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W37-2012

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W38-2012
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W40-2012
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W41-2012
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W44-2012
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W50-2012
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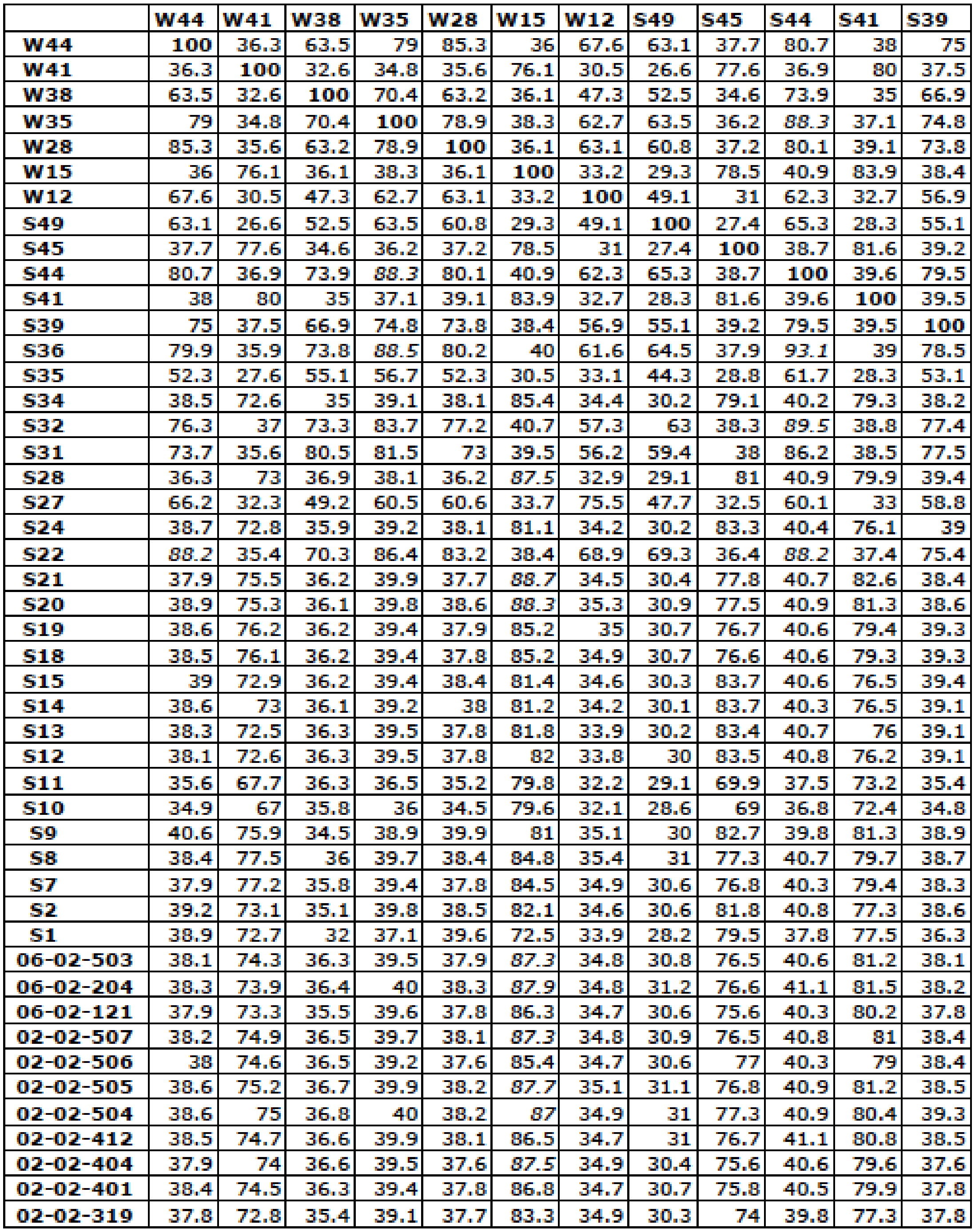
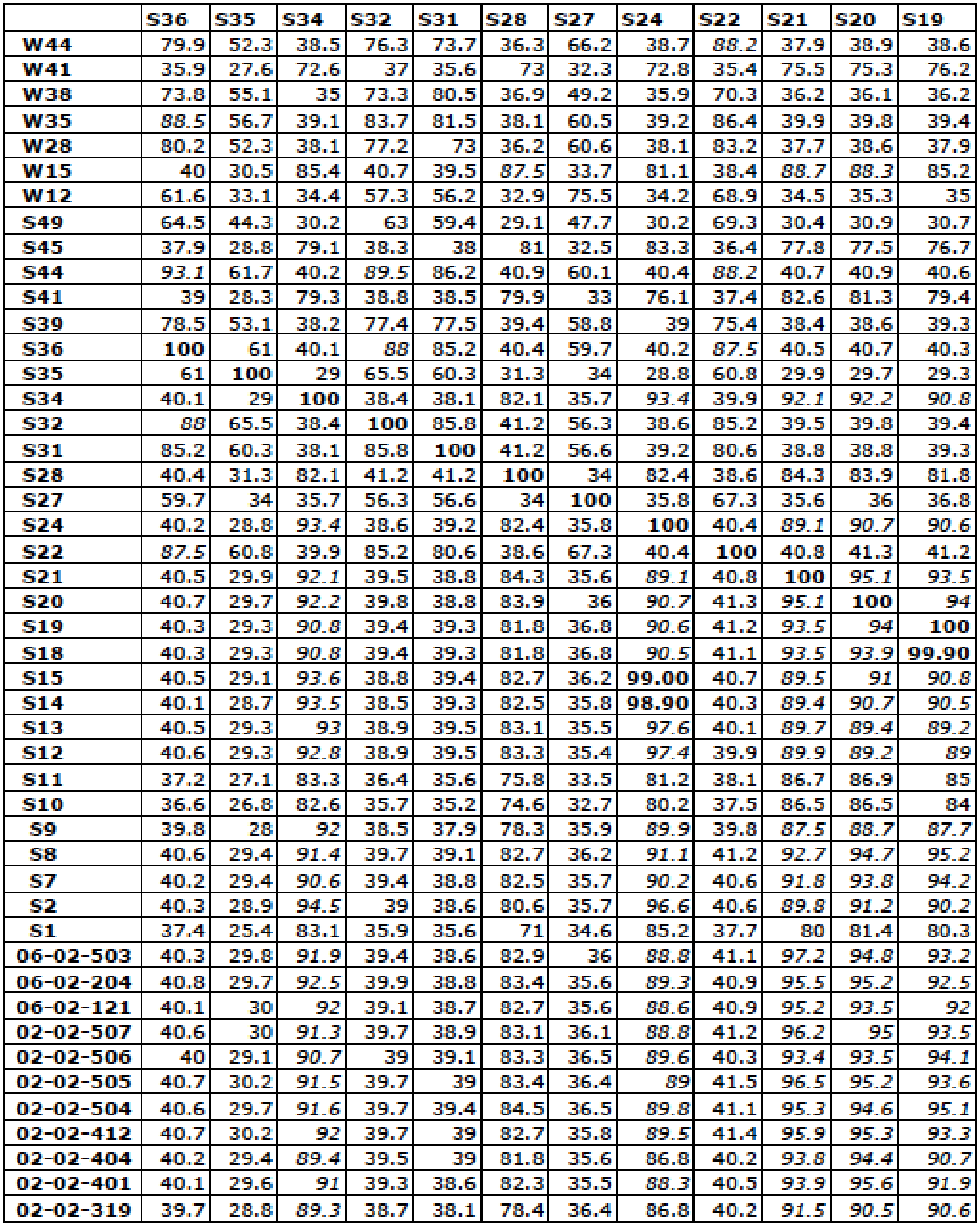
TTAGCTGAGAGGGGGGGCTTGGCCCCTCTCCATT

Tables

Table A1: Complete similarity matrix for summer clones compared to winter clones. Italicized numbers indicate a similarity index of 87 or greater; bolded numbers indicate sequences with a similarity index of 98.7 or greater.

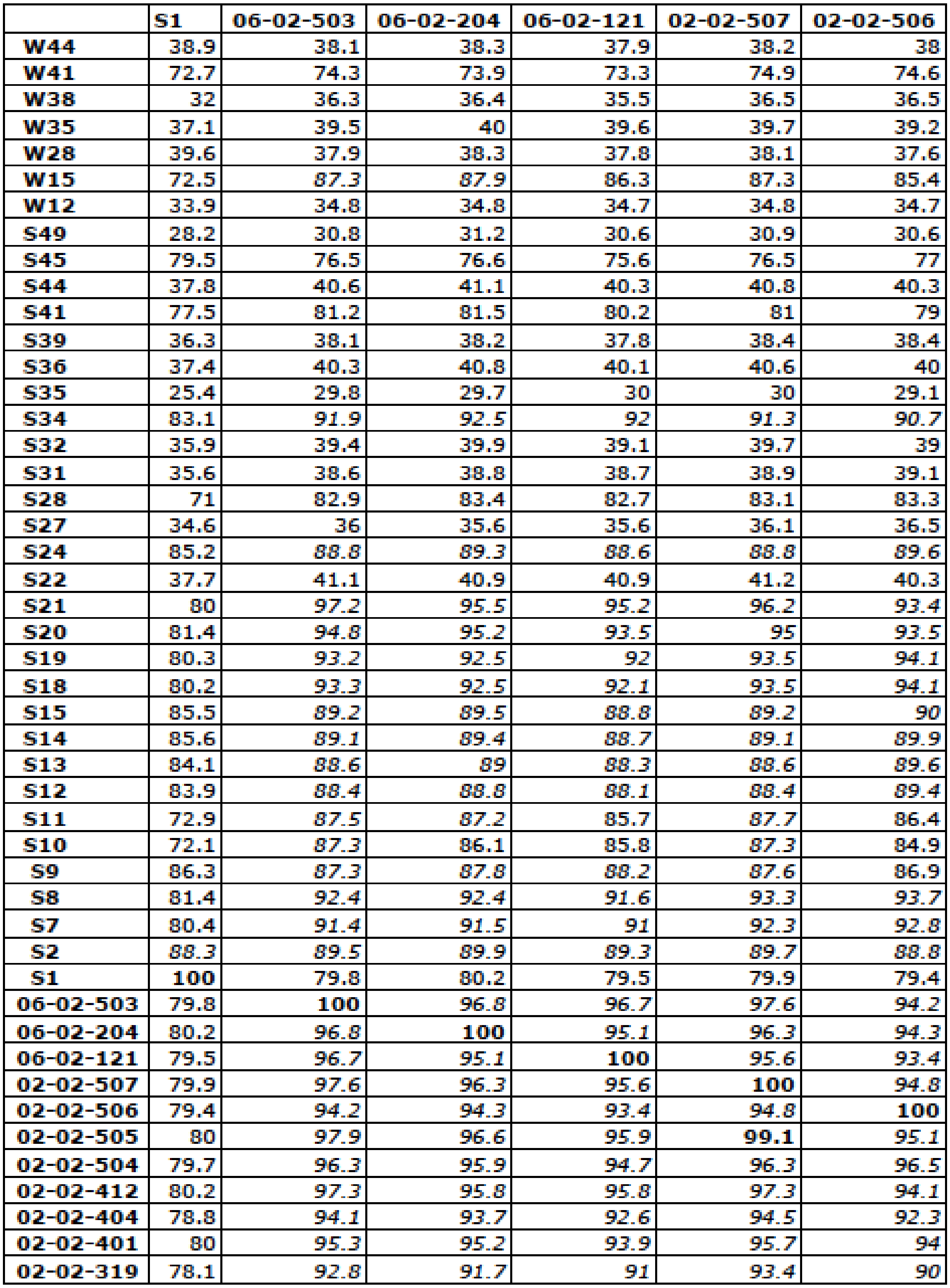


	W44	W41	W38	W35	W28	W15	W12	S49	S45	S44	S41	S39
W44	**100**	36.3	63.5	79	85.3	36	67.6	63.1	37.7	80.7	38	75
W41	36.3	**100**	32.6	34.8	35.6	76.1	30.5	26.6	77.6	36.9	80	37.5
W38	63.5	32.6	**100**	70.4	63.2	36.1	47.3	52.5	34.6	73.9	35	66.9
W35	79	34.8	70.4	**100**	78.9	38.3	62.7	63.5	36.2	*88.3*	37.1	74.8
W28	85.3	35.6	63.2	78.9	**100**	36.1	63.1	60.8	37.2	80.1	39.1	73.8
W15	36	76.1	36.1	38.3	36.1	**100**	33.2	29.3	78.5	40.9	83.9	38.4
W12	67.6	30.5	47.3	62.7	63.1	33.2	**100**	49.1	31	62.3	32.7	56.9
S49	63.1	26.6	52.5	63.5	60.8	29.3	49.1	**100**	27.4	65.3	28.3	55.1
S45	37.7	77.6	34.6	36.2	37.2	78.5	31	27.4	**100**	38.7	81.6	39.2
S44	80.7	36.9	73.9	*88.3*	80.1	40.9	62.3	65.3	38.7	**100**	39.6	79.5
S41	38	80	35	37.1	39.1	83.9	32.7	28.3	81.6	39.6	**100**	39.5
S39	75	37.5	66.9	74.8	73.8	38.4	56.9	55.1	39.2	79.5	39.5	**100**
S36	79.9	35.9	73.8	*88.5*	80.2	40	61.6	64.5	37.9	*93.1*	39	78.5
S35	52.3	27.6	55.1	56.7	52.3	30.5	33.1	44.3	28.8	61.7	28.3	53.1
S34	38.5	72.6	35	39.1	38.1	85.4	34.4	30.2	79.1	40.2	79.3	38.2
S32	76.3	37	73.3	83.7	77.2	40.7	57.3	63	38.3	*89.5*	38.8	77.4
S31	73.7	35.6	80.5	81.5	73	39.5	56.2	59.4	38	86.2	38.5	77.5
S28	36.3	73	36.9	38.1	36.2	*87.5*	32.9	29.1	81	40.9	79.9	39.4
S27	66.2	32.3	49.2	60.5	60.6	33.7	75.5	47.7	32.5	60.1	33	58.8
S24	38.7	72.8	35.9	39.2	38.1	81.1	34.2	30.2	83.3	40.4	76.1	39
S22	*88.2*	35.4	70.3	86.4	83.2	38.4	68.9	69.3	36.4	*88.2*	37.4	75.4
S21	37.9	75.5	36.2	39.9	37.7	*88.7*	34.5	30.4	77.8	40.7	82.6	38.4
S20	38.9	75.3	36.1	39.8	38.6	*88.3*	35.3	30.9	77.5	40.9	81.3	38.6
S19	38.6	76.2	36.2	39.4	37.9	85.2	35	30.7	76.7	40.6	79.4	39.3
S18	38.5	76.1	36.2	39.4	37.8	85.2	34.9	30.7	76.6	40.6	79.3	39.3
S15	39	72.9	36.2	39.4	38.4	81.4	34.6	30.3	83.7	40.6	76.5	39.4
S14	38.6	73	36.1	39.2	38	81.2	34.2	30.1	83.7	40.3	76.5	39.1
S13	38.3	72.5	36.3	39.5	37.8	81.8	33.9	30.2	83.4	40.7	76	39.1
S12	38.1	72.6	36.3	39.5	37.8	82	33.8	30	83.5	40.8	76.2	39.1
S11	35.6	67.7	36.3	36.5	35.2	79.8	32.2	29.1	69.9	37.5	73.2	35.4
S10	34.9	67	35.8	36	34.5	79.6	32.1	28.6	69	36.8	72.4	34.8
S9	40.6	75.9	34.5	38.9	39.9	81	35.1	30	82.7	39.8	81.3	38.9
S8	38.4	77.5	36	39.7	38.4	84.8	35.4	31	77.3	40.7	79.7	38.7
S7	37.9	77.2	35.8	39.4	37.8	84.5	34.9	30.6	76.8	40.3	79.4	38.3
S2	39.2	73.1	35.1	39.8	38.5	82.1	34.6	30.6	81.8	40.8	77.3	38.6
S1	38.9	72.7	32	37.1	39.6	72.5	33.9	28.2	79.5	37.8	77.5	36.3
06-02-503	38.1	74.3	36.3	39.5	37.9	*87.3*	34.8	30.8	76.5	40.6	81.2	38.1
06-02-204	38.3	73.9	36.4	40	38.3	*87.9*	34.8	31.2	76.6	41.1	81.5	38.2
06-02-121	37.9	73.3	35.5	39.6	37.8	86.3	34.7	30.6	75.6	40.3	80.2	37.8
02-02-507	38.2	74.9	36.5	39.7	38.1	*87.3*	34.8	30.9	76.5	40.8	81	38.4
02-02-506	38	74.6	36.5	39.2	37.6	85.4	34.7	30.6	77	40.3	79	38.4
02-02-505	38.6	75.2	36.7	39.9	38.2	*87.7*	35.1	31.1	76.8	40.9	81.2	38.5
02-02-504	38.6	75	36.8	40	38.2	*87*	34.9	31	77.3	40.9	80.4	39.3
02-02-412	38.5	74.7	36.6	39.9	38.1	86.5	34.7	31	76.7	41.1	80.8	38.5
02-02-404	37.9	74	36.6	39.5	37.6	*87.5*	34.9	30.4	75.6	40.6	79.6	37.6
02-02-401	38.4	74.5	36.3	39.4	37.8	86.8	34.7	30.7	75.8	40.5	79.9	37.8
02-02-319	37.8	72.8	35.4	39.1	37.7	83.3	34.9	30.3	74	39.8	77.3	37.8



	S36	**S35**	**S34**	**S32**	**S31**	**S28**	**S27**	**S24**	**S22**	**S21**	**S20**	**S19**
W44	79.9	52.3	38.5	76.3	73.7	36.3	66.2	38.7	*88.2*	37.9	38.9	38.6
W41	35.9	27.6	72.6	37	35.6	73	32.3	72.8	35.4	75.5	75.3	76.2
W38	73.8	55.1	35	73.3	80.5	36.9	49.2	35.9	70.3	36.2	36.1	36.2
W35	*88.5*	56.7	39.1	83.7	81.5	38.1	60.5	39.2	86.4	39.9	39.8	39.4
W28	80.2	52.3	38.1	77.2	73	36.2	60.6	38.1	83.2	37.7	38.6	37.9
W15	40	30.5	85.4	40.7	39.5	*87.5*	33.7	81.1	38.4	*88.7*	*88.3*	85.2
W12	61.6	33.1	34.4	57.3	56.2	32.9	75.5	34.2	68.9	34.5	35.3	35
S49	64.5	44.3	30.2	63	59.4	29.1	47.7	30.2	69.3	30.4	30.9	30.7
S45	37.9	28.8	79.1	38.3	38	81	32.5	83.3	36.4	77.8	77.5	76.7
S44	*93.1*	61.7	40.2	*89.5*	86.2	40.9	60.1	40.4	*88.2*	40.7	40.9	40.6
S41	39	28.3	79.3	38.8	38.5	79.9	33	76.1	37.4	82.6	81.3	79.4
S39	78.5	53.1	38.2	77.4	77.5	39.4	58.8	39	75.4	38.4	38.6	39.3
S36	**100**	61	40.1	*88*	85.2	40.4	59.7	40.2	*87.5*	40.5	40.7	40.3
S35	61	**100**	29	65.5	60.3	31.3	34	28.8	60.8	29.9	29.7	29.3
S34	40.1	29	**100**	38.4	38.1	82.1	35.7	*93.4*	39.9	*92.1*	*92.2*	*90.8*
S32	*88*	65.5	38.4	**100**	85.8	41.2	56.3	38.6	85.2	39.5	39.8	39.4
S31	85.2	60.3	38.1	85.8	**100**	41.2	56.6	39.2	80.6	38.8	38.8	39.3
S28	40.4	31.3	82.1	41.2	41.2	**100**	34	82.4	38.6	84.3	83.9	81.8
S27	59.7	34	35.7	56.3	56.6	34	**100**	35.8	67.3	35.6	36	36.8
S24	40.2	28.8	*93.4*	38.6	39.2	82.4	35.8	**100**	40.4	*89.1*	*90.7*	*90.6*
S22	*87.5*	60.8	39.9	85.2	80.6	38.6	67.3	40.4	**100**	40.8	41.3	41.2
S21	40.5	29.9	*92.1*	39.5	38.8	84.3	35.6	*89.1*	40.8	**100**	*95.1*	*93.5*
S20	40.7	29.7	*92.2*	39.8	38.8	83.9	36	*90.7*	41.3	*95.1*	**100**	*94*
S19	40.3	29.3	*90.8*	39.4	39.3	81.8	36.8	*90.6*	41.2	*93.5*	*94*	**100**
S18	40.3	29.3	*90.8*	39.4	39.3	81.8	36.8	*90.5*	41.1	*93.5*	*93.9*	**99.90**
S15	40.5	29.1	*93.6*	38.8	39.4	82.7	36.2	**99.00**	40.7	*89.5*	*91*	*90.8*
S14	40.1	28.7	*93.5*	38.5	39.3	82.5	35.8	**98.90**	40.3	*89.4*	*90.7*	*90.5*
S13	40.5	29.3	*93*	38.9	39.5	83.1	35.5	*97.6*	40.1	*89.7*	*89.4*	*89.2*
S12	40.6	29.3	*92.8*	38.9	39.5	83.3	35.4	*97.4*	39.9	*89.9*	*89.2*	*89*
S11	37.2	27.1	83.3	36.4	35.6	75.8	33.5	81.2	38.1	86.7	86.9	85
S10	36.6	26.8	82.6	35.7	35.2	74.6	32.7	80.2	37.5	86.5	86.5	84
S9	39.8	28	*92*	38.5	37.9	78.3	35.9	*89.9*	39.8	*87.5*	*88.7*	*87.7*
S8	40.6	29.4	*91.4*	39.7	39.1	82.7	36.2	*91.1*	41.2	*92.7*	*94.7*	*95.2*
S7	40.2	29.4	*90.6*	39.4	38.8	82.5	35.7	*90.2*	40.6	*91.8*	*93.8*	*94.2*
S2	40.3	28.9	*94.5*	39	38.6	80.6	35.7	96.6	40.6	*89.8*	*91.2*	*90.2*
S1	37.4	25.4	83.1	35.9	35.6	71	34.6	85.2	37.7	80	81.4	80.3
06-02-503	40.3	29.8	*91.9*	39.4	38.6	82.9	36	*88.8*	41.1	*97.2*	*94.8*	*93.2*
06-02-204	40.8	29.7	*92.5*	39.9	38.8	83.4	35.6	*89.3*	40.9	*95.5*	*95.2*	*92.5*
06-02-121	40.1	30	*92*	39.1	38.7	82.7	35.6	*88.6*	40.9	*95.2*	*93.5*	*92*
02-02-507	40.6	30	*91.3*	39.7	38.9	83.1	36.1	*88.8*	41.2	*96.2*	*95*	*93.5*
02-02-506	40	29.1	*90.7*	39	39.1	83.3	36.5	*89.6*	40.3	*93.4*	*93.5*	*94.1*
02-02-505	40.7	30.2	*91.5*	39.7	39	83.4	36.4	*89*	41.5	*96.5*	*95.2*	*93.6*
02-02-504	40.6	29.7	*91.6*	39.7	39.4	84.5	36.5	*89.8*	41.1	*95.3*	*94.6*	*95.1*
02-02-412	40.7	30.2	*92*	39.7	39	82.7	35.8	*89.5*	41.4	*95.9*	*95.3*	*93.3*
02-02-404	40.2	29.4	*89.4*	39.5	39	81.8	35.6	86.8	40.2	*93.8*	*94.4*	*90.7*
02-02-401	40.1	29.6	*91*	39.3	38.6	82.3	35.5	*88.3*	40.5	*93.9*	*95.6*	*91.9*
02-02-319	39.7	28.8	*89.3*	38.7	38.1	78.4	36.4	86.8	40.2	*91.5*	*90.5*	*90.6*

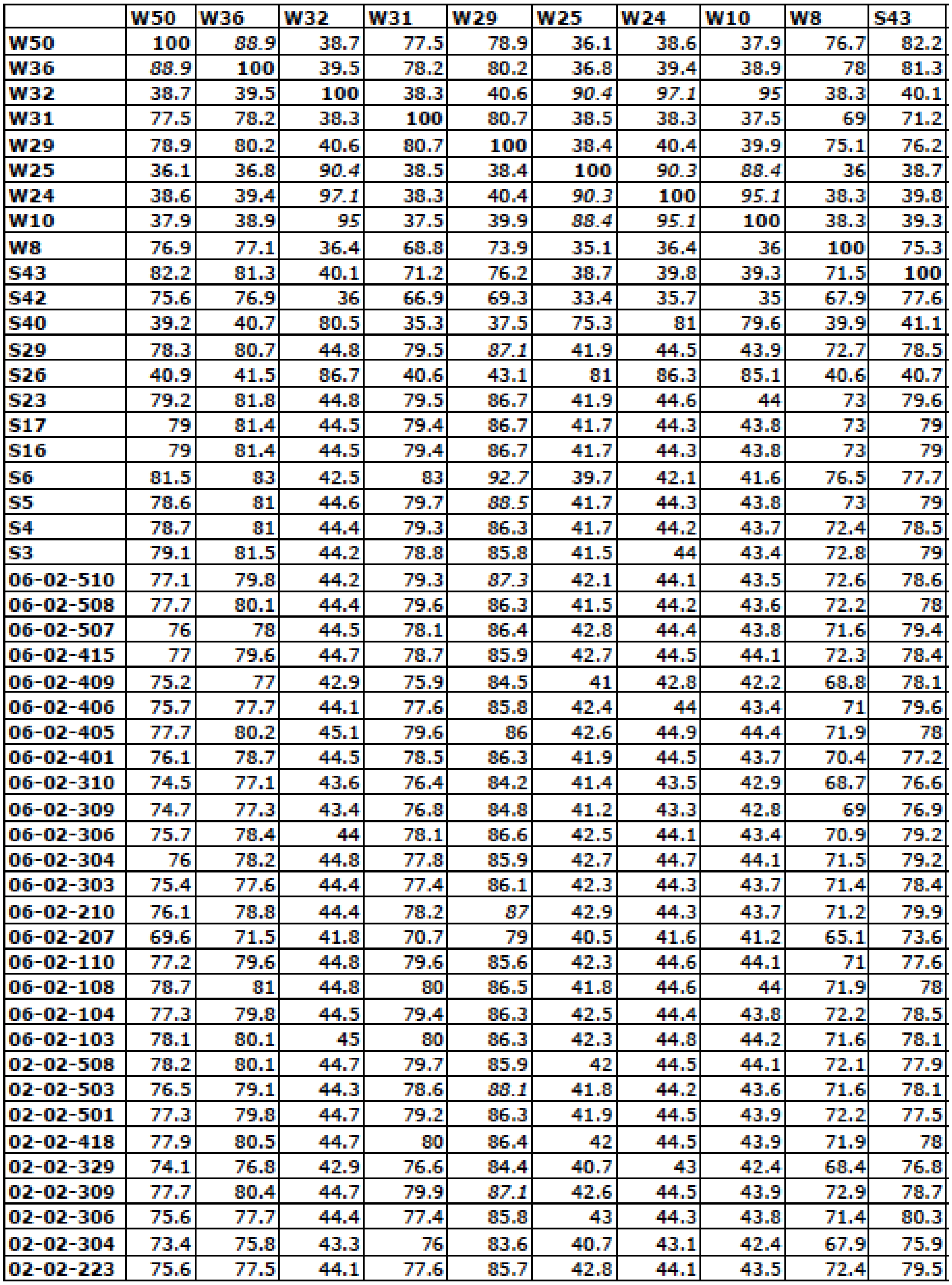
	S18	**S15**	**S14**	**S13**	**S12**	**S11**	**S10**	**S9**	**S8**	**S7**	**S2**
W44	38.5	39	38.6	38.3	38.1	35.6	34.9	40.6	38.4	37.9	39.2
W41	76.1	72.9	73	72.5	72.6	67.7	67	75.9	77.5	77.2	73.1
W38	36.2	36.2	36.1	36.3	36.3	36.3	35.8	34.5	36	35.8	35.1
W35	39.4	39.4	39.2	39.5	39.5	36.5	36	38.9	39.7	39.4	39.8
W28	37.8	38.4	38	37.8	37.8	35.2	34.5	39.9	38.4	37.8	38.5
W15	85.2	81.4	81.2	81.8	82	79.8	79.6	81	84.8	84.5	82.1
W12	34.9	34.6	34.2	33.9	33.8	32.2	32.1	35.1	35.4	34.9	34.6
S49	30.7	30.3	30.1	30.2	30	29.1	28.6	30	31	30.6	30.6
S45	76.6	83.7	83.7	83.4	83.5	69.9	69	82.7	77.3	76.8	81.8
S44	40.6	40.6	40.3	40.7	40.8	37.5	36.8	39.8	40.7	40.3	40.8
S41	79.3	76.5	76.5	76	76.2	73.2	72.4	81.3	79.7	79.4	77.3
S39	39.3	39.4	39.1	39.1	39.1	35.4	34.8	38.9	38.7	38.3	38.6
S36	40.3	40.5	40.1	40.5	40.6	37.2	36.6	39.8	40.6	40.2	40.3
S35	29.3	29.1	28.7	29.3	29.3	27.1	26.8	28	29.4	29.4	28.9
S34	*90.8*	*93.6*	*93.5*	*93*	*92.8*	83.3	82.6	*92*	91.4	90.6	94.5
S32	39.4	38.8	38.5	38.9	38.9	36.4	35.7	38.5	39.7	39.4	39
S31	39.3	39.4	39.3	39.5	39.5	35.6	35.2	37.9	39.1	38.8	38.6
S28	81.8	82.7	82.5	83.1	83.3	75.8	74.6	78.3	82.7	82.5	80.6
S27	36.8	36.2	35.8	35.5	35.4	33.5	32.7	35.9	36.2	35.7	35.7
S24	*90.5*	**99.00**	**98.9**	*97.6*	*97.4*	81.2	80.2	*89.9*	*91.1*	*90.2*	*96.6*
S22	41.1	40.7	40.3	40.1	39.9	38.1	37.5	39.8	41.2	40.6	40.6
S21	*93.5*	*89.5*	*89.4*	*89.7*	*89.9*	86.7	86.5	*87.5*	*92.7*	*91.8*	*89.8*
S20	*93.9*	*91*	*90.7*	*89.4*	*89.2*	86.9	86.5	*88.7*	*94.7*	*93.8*	*91.2*
S19	**99.90**	*90.8*	*90.5*	*89.2*	*89*	85	84	*87.7*	*95.2*	*94.2*	*90.2*
S18	**100**	*90.7*	*90.4*	*89.2*	*89*	85	84	*87.6*	*95.1*	*94.1*	*90.1*
S15	*90.7*	**100**	**99.10**	*97.8*	*97.6*	81.5	80.6	*90.1*	*91.2*	*90.4*	*96.8*
S14	*90.4*	99.1	**100**	*97.7*	*97.5*	81.4	80.5	*90.3*	*91.1*	*90.4*	*96.9*
S13	*89.2*	97.8	*97.7*	**100**	**99.80**	80.6	79.7	*88.8*	*89.7*	*88.8*	*95.4*
S12	*89*	97.6	*97.5*	**99.80**	**100**	80.5	79.5	*88.6*	*89.5*	*88.6*	*95.2*
S11	85	81.5	81.4	80.6	80.5	**100**	*97.1*	79.8	84.6	83.8	81.9
S10	84	80.6	80.5	79.7	79.5	*97.1*	**100**	78.9	83.8	82.9	81.1
S9	*87.6*	*90.1*	*90.3*	*88.8*	*88.6*	79.8	78.9	**100**	*88.1*	*87.1*	*91.2*
S8	*95.1*	*91.2*	*91.1*	*89.7*	*89.5*	84.6	83.8	*88.1*	**100**	***98.80***	*91.2*
S7	*94.1*	*90.4*	*90.4*	*88.8*	*88.6*	83.8	82.9	*87.1*	**98.80**	**100**	*90.2*
S2	*90.1*	*96.8*	*96.9*	*95.4*	*95.2*	81.9	81.1	*91.2*	*91.2*	*90.2*	**100**
S1	80.2	85.5	85.6	84.1	83.9	72.9	72.1	86.3	81.4	80.4	*88.3*
06-02-503	*93.3*	*89.2*	*89.1*	*88.6*	*88.4*	*87.5*	*87.3*	*87.3*	92.4	*91.4*	*89.5*
06-02-204	*92.5*	*89.5*	*89.4*	*89*	*88.8*	*87.2*	86.1	*87.8*	92.4	*91.5*	*89.9*
06-02-121	*92.1*	*88.8*	*88.7*	*88.3*	*88.1*	85.7	85.8	*88.2*	91.6	*91*	*89.3*
02-02-507	*93.5*	*89.2*	*89.1*	*88.6*	*88.4*	*87.7*	*87.3*	*87.6*	*93.3*	*92.3*	*89.7*
02-02-506	*94.1*	*90*	*89.9*	*89.6*	*89.4*	86.4	84.9	86.9	*93.7*	*92.8*	*88.8*
02-02-505	*93.7*	*89.4*	*89.3*	*88.8*	*88.6*	*88*	*87.6*	*87.8*	*93.5*	*92.6*	*89.8*
02-02-504	*95.2*	*90.1*	*90*	*89.5*	*89.3*	*87.1*	86.2	*87.2*	*93.2*	*92.2*	*89.6*
02-02-412	*93.3*	*89.9*	*89.6*	*89.5*	*89.3*	86.9	86.7	*87.7*	*93.3*	*92.3*	*90.2*
02-02-404	*90.8*	*87.1*	*87*	*87.3*	*87.3*	85.6	85.7	85.4	*91*	*90*	*88*
02-02-401	*92*	*88.5*	*88.3*	*87.8*	*87.7*	86.9	86.2	*87.1*	*92.5*	*91.6*	*89.3*
02-02-319	*90.6*	*87*	86.7	86.5	86.3	83.4	83.3	85.3	*89.9*	*89*	*87.8*



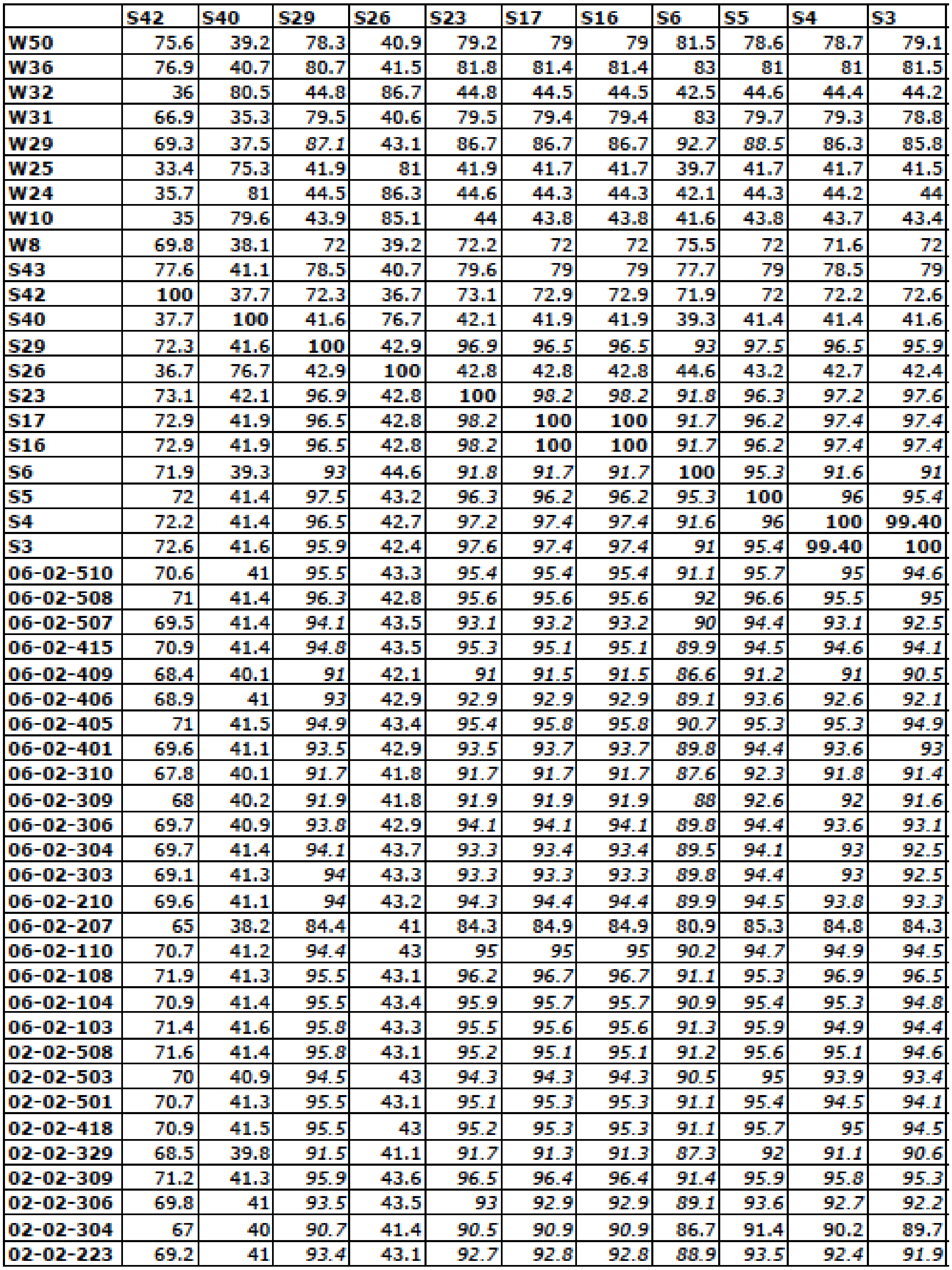
	S1	**06-02-503**	**06-02-204**	**06-02-121**	**02-02-507**	**02-02-506**
W44	38.9	38.1	38.3	37.9	38.2	38
W41	72.7	74.3	73.9	73.3	74.9	74.6
W38	32	36.3	36.4	35.5	36.5	36.5
W35	37.1	39.5	40	39.6	39.7	39.2
W28	39.6	37.9	38.3	37.8	38.1	37.6
W15	72.5	*87.3*	*87.9*	86.3	87.3	85.4
W12	33.9	34.8	34.8	34.7	34.8	34.7
S49	28.2	30.8	31.2	30.6	30.9	30.6
S45	79.5	76.5	76.6	75.6	76.5	77
S44	37.8	40.6	41.1	40.3	40.8	40.3
S41	77.5	81.2	81.5	80.2	81	79
S39	36.3	38.1	38.2	37.8	38.4	38.4
S36	37.4	40.3	40.8	40.1	40.6	40
S35	25.4	29.8	29.7	30	30	29.1
S34	83.1	*91.9*	*92.5*	*92*	*91.3*	*90.7*
S32	35.9	39.4	39.9	39.1	39.7	39
S31	35.6	38.6	38.8	38.7	38.9	39.1
S28	71	82.9	83.4	82.7	83.1	83.3
S27	34.6	36	35.6	35.6	36.1	36.5
S24	85.2	*88.8*	*89.3*	*88.6*	*88.8*	*89.6*
S22	37.7	41.1	40.9	40.9	41.2	40.3
S21	80	*97.2*	*95.5*	*95.2*	*96.2*	*93.4*
S20	81.4	*94.8*	*95.2*	*93.5*	*95*	*93.5*
S19	80.3	*93.2*	*92.5*	*92*	*93.5*	*94.1*
S18	80.2	*93.3*	*92.5*	*92.1*	*93.5*	*94.1*
S15	85.5	*89.2*	*89.5*	*88.8*	*89.2*	*90*
S14	85.6	*89.1*	*89.4*	*88.7*	*89.1*	*89.9*
S13	84.1	*88.6*	*89*	*88.3*	*88.6*	*89.6*
S12	83.9	*88.4*	*88.8*	*88.1*	*88.4*	*89.4*
S11	72.9	*87.5*	*87.2*	85.7	*87.7*	86.4
S10	72.1	*87.3*	86.1	85.8	*87.3*	84.9
S9	86.3	*87.3*	*87.8*	*88.2*	*87.6*	86.9
S8	81.4	*92.4*	*92.4*	*91.6*	*93.3*	*93.7*
S7	80.4	*91.4*	*91.5*	*91*	*92.3*	*92.8*
S2	*88.3*	*89.5*	*89.9*	*89.3*	*89.7*	*88.8*
S1	**100**	79.8	80.2	79.5	79.9	79.4
06-02-503	79.8	**100**	*96.8*	*96.7*	*97.6*	*94.2*
06-02-204	80.2	*96.8*	**100**	*95.1*	*96.3*	*94.3*
06-02-121	79.5	*96.7*	*95.1*	**100**	*95.6*	*93.4*
02-02-507	79.9	*97.6*	*96.3*	*95.6*	**100**	*94.8*
02-02-506	79.4	*94.2*	*94.3*	*93.4*	*94.8*	**100**
02-02-505	80	*97.9*	*96.6*	*95.9*	**99.1**	*95.1*
02-02-504	79.7	*96.3*	*95.9*	*94.7*	*96.3*	*96.5*
02-02-412	80.2	*97.3*	*95.8*	*95.8*	*97.3*	*94.1*
02-02-404	78.8	*94.1*	*93.7*	*92.6*	*94.5*	*92.3*
02-02-401	80	*95.3*	*95.2*	*93.9*	*95.7*	*94*
02-02-319	78.1	*92.8*	*91.7*	*91*	*93.4*	*90*

	02-02-505	**02-02-504**	**02-02-412**	**02-02-404**	**02-02-401**	**02-02-319**
W44	38.6	38.6	38.5	37.9	38.4	37.8
W41	75.2	75	74.7	74	74.5	72.8
W38	36.7	36.8	36.6	36.6	36.3	35.4
W35	39.9	40	39.9	39.5	39.4	39.1
W28	38.2	38.2	38.1	37.6	37.8	37.7
W15	87.7	87	86.5	87.5	86.8	83.3
W12	35.1	34.9	34.7	34.9	34.7	34.9
S49	31.1	31	31	30.4	30.7	30.3
S45	76.8	77.3	76.7	75.6	75.8	74
S44	40.9	40.9	41.1	40.6	40.5	39.8
S41	81.2	80.4	80.8	79.6	79.9	77.3
S39	38.5	39.3	38.5	37.6	37.8	37.8
S36	40.7	40.6	40.7	40.2	40.1	39.7
S35	30.2	29.7	30.2	29.4	29.6	28.8
S34	*91.5*	*91.6*	*92*	*89.4*	*91*	*89.3*
S32	39.7	39.7	39.7	39.5	39.3	38.7
S31	39	39.4	39	39	38.6	38.1
S28	83.4	84.5	82.7	81.8	82.3	78.4
S27	36.4	36.5	35.8	35.6	35.5	36.4
S24	*89*	*89.8*	*89.5*	86.8	*88.3*	86.8
S22	41.5	41.1	41.4	40.2	40.5	40.2
S21	*96.5*	*95.3*	*95.9*	*93.8*	*93.9*	*91.5*
S20	*95.2*	*94.6*	*95.3*	*94.4*	*95.6*	*90.5*
S19	*93.6*	*95.1*	*93.3*	*90.7*	*91.9*	*90.6*
S18	*93.7*	*95.2*	*93.3*	*90.8*	*92*	*90.6*
S15	*89.4*	*90.1*	*89.9*	*87.1*	*88.5*	*87*
S14	*89.3*	*90*	*89.6*	*87*	*88.3*	86.7
S13	*88.8*	*89.5*	*89.5*	*87.3*	*87.8*	86.5
S12	*88.6*	*89.3*	*89.3*	*87.3*	*87.7*	86.3
S11	*88*	*87.1*	86.9	85.6	86.9	83.4
S10	*87.6*	86.2	86.7	85.7	86.2	83.3
S9	*87.8*	*87.2*	*87.7*	85.4	*87.1*	85.3
S8	*93.5*	*93.2*	*93.3*	*91*	*92.5*	*89.9*
S7	*92.6*	*92.2*	*92.3*	*90*	*91.6*	*89*
S2	*89.8*	*89.6*	*90.2*	*88*	*89.3*	*87.8*
S1	80	79.7	80.2	78.8	80	78.1
06-02-503	*97.9*	*96.3*	*97.3*	*94.1*	*95.3*	*92.8*
06-02-204	*96.6*	*95.9*	*95.8*	*93.7*	*95.2*	*91.7*
06-02-121	*95.9*	*94.7*	*95.8*	*92.6*	*93.9*	*91*
02-02-507	**99.1**	*96.3*	*97.3*	*94.5*	*95.7*	*93.4*
02-02-506	*95.1*	*96.5*	*94.1*	*92.3*	*94*	*90*
02-02-505	**100**	*96.6*	*97.6*	*94.8*	*96.2*	*93.7*
02-02-504	*96.6*	**100**	*95.5*	*93.3*	*94.7*	*91.3*
02-02-412	*97.6*	*95.5*	**100**	*94.4*	*95.6*	*92.7*
02-02-404	*94.8*	*93.3*	*94.4*	**100**	*96*	*90.4*
02-02-401	*96.2*	*94.7*	*95.6*	*96*	**100**	*91.2*
02-02-319	*93.7*	*91.3*	*92.7*	*90.4*	*91.2*	**100**

Table A2: Complete similarity matrix for summer and winter clones that were most closely related to *Methanosaeta concilii* compared to Grosser's clones that were most closely related to *Methanosaeta concilii*. Italicized numbers indicate a similarity index of 87 or greater; bolded numbers indicate sequences with a similarity index of 98.7 or greater.

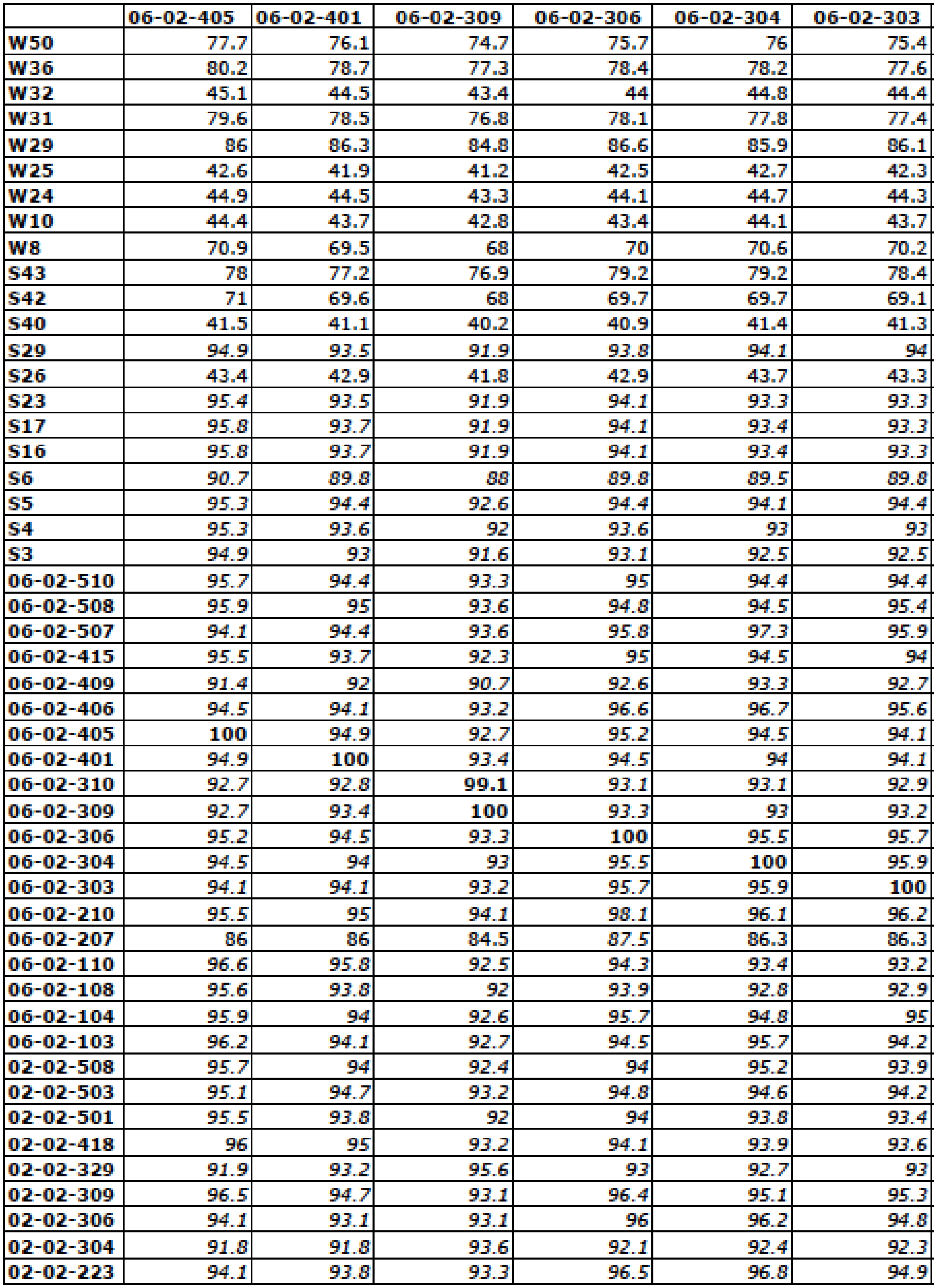


	W50	W36	W32	W31	W29	W25	W24	W10	W8	S43
W50	**100**	*88.9*	38.7	77.5	78.9	36.1	38.6	37.9	76.7	82.2
W36	*88.9*	**100**	39.5	78.2	80.2	36.8	39.4	38.9	78	81.3
W32	38.7	39.5	**100**	38.3	40.6	*90.4*	*97.1*	*95*	38.3	40.1
W31	77.5	78.2	38.3	**100**	80.7	38.5	38.3	37.5	69	71.2
W29	78.9	80.2	40.6	80.7	**100**	38.4	40.4	39.9	75.1	76.2
W25	36.1	36.8	*90.4*	38.5	38.4	**100**	*90.3*	*88.4*	36	38.7
W24	38.6	39.4	*97.1*	38.3	40.4	*90.3*	**100**	*95.1*	38.3	39.8
W10	37.9	38.9	*95*	37.5	39.9	*88.4*	*95.1*	**100**	38.3	39.3
W8	76.9	77.1	36.4	68.8	73.9	35.1	36.4	36	**100**	75.3
S43	82.2	81.3	40.1	71.2	76.2	38.7	39.8	39.3	71.5	**100**
S42	75.6	76.9	36	66.9	69.3	33.4	35.7	35	67.9	77.6
S40	39.2	40.7	80.5	35.3	37.5	75.3	81	79.6	39.9	41.1
S29	78.3	80.7	44.8	79.5	*87.1*	41.9	44.5	43.9	72.7	78.5
S26	40.9	41.5	86.7	40.6	43.1	81	86.3	85.1	40.6	40.7
S23	79.2	81.8	44.8	79.5	86.7	41.9	44.6	44	73	79.6
S17	79	81.4	44.5	79.4	86.7	41.7	44.3	43.8	73	79
S16	79	81.4	44.5	79.4	86.7	41.7	44.3	43.8	73	79
S6	81.5	83	42.5	83	*92.7*	39.7	42.1	41.6	76.5	77.7
S5	78.6	81	44.6	79.7	*88.5*	41.7	44.3	43.8	73	79
S4	78.7	81	44.4	79.3	86.3	41.7	44.2	43.7	72.4	78.5
S3	79.1	81.5	44.2	78.8	85.8	41.5	44	43.4	72.8	79
06-02-510	77.1	79.8	44.2	79.3	*87.3*	42.1	44.1	43.5	72.6	78.6
06-02-508	77.7	80.1	44.4	79.6	86.3	41.5	44.2	43.6	72.2	78
06-02-507	76	78	44.5	78.1	86.4	42.8	44.4	43.8	71.6	79.4
06-02-415	77	79.6	44.7	78.7	85.9	42.7	44.5	44.1	72.3	78.4
06-02-409	75.2	77	42.9	75.9	84.5	41	42.8	42.2	68.8	78.1
06-02-406	75.7	77.7	44.1	77.6	85.8	42.4	44	43.4	71	79.6
06-02-405	77.7	80.2	45.1	79.6	86	42.6	44.9	44.4	71.9	78
06-02-401	76.1	78.7	44.5	78.5	86.3	41.9	44.5	43.7	70.4	77.2
06-02-310	74.5	77.1	43.6	76.4	84.2	41.4	43.5	42.9	68.7	76.6
06-02-309	74.7	77.3	43.4	76.8	84.8	41.2	43.3	42.8	69	76.9
06-02-306	75.7	78.4	44	78.1	86.6	42.5	44.1	43.4	70.9	79.2
06-02-304	76	78.2	44.8	77.8	85.9	42.7	44.7	44.1	71.5	79.2
06-02-303	75.4	77.6	44.4	77.4	86.1	42.3	44.3	43.7	71.4	78.4
06-02-210	76.1	78.8	44.4	78.2	*87*	42.9	44.3	43.7	71.2	79.9
06-02-207	69.6	71.5	41.8	70.7	79	40.5	41.6	41.2	65.1	73.6
06-02-110	77.2	79.6	44.8	79.6	85.6	42.3	44.6	44.1	71	77.6
06-02-108	78.7	81	44.8	80	86.5	41.8	44.6	44	71.9	78
06-02-104	77.3	79.8	44.5	79.4	86.3	42.5	44.4	43.8	72.2	78.5
06-02-103	78.1	80.1	45	80	86.3	42.3	44.8	44.2	71.6	78.1
02-02-508	78.2	80.1	44.7	79.7	85.9	42	44.5	44.1	72.1	77.9
02-02-503	76.5	79.1	44.3	78.6	*88.1*	41.8	44.2	43.6	71.6	78.1
02-02-501	77.3	79.8	44.7	79.2	86.3	41.9	44.5	43.9	72.2	77.5
02-02-418	77.9	80.5	44.7	80	86.4	42	44.5	43.9	71.9	78
02-02-329	74.1	76.8	42.9	76.6	84.4	40.7	43	42.4	68.4	76.8
02-02-309	77.7	80.4	44.7	79.9	*87.1*	42.6	44.5	43.9	72.9	78.7
02-02-306	75.6	77.7	44.4	77.4	85.8	43	44.3	43.8	71.4	80.3
02-02-304	73.4	75.8	43.3	76	83.6	40.7	43.1	42.4	67.9	75.9
02-02-223	75.6	77.5	44.1	77.6	85.7	42.8	44.1	43.5	72.4	79.5



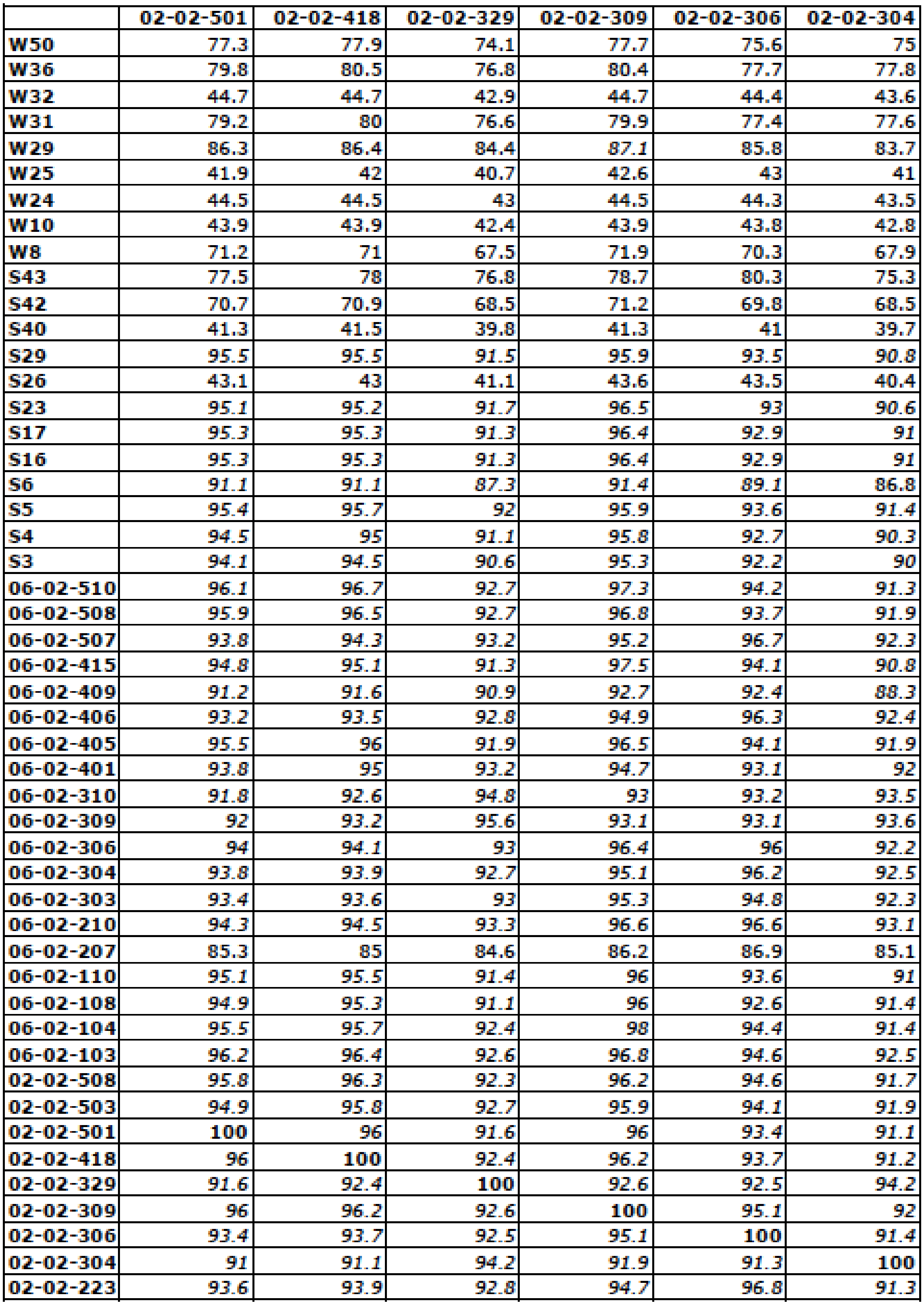
	S42	S40	S29	S26	S23	S17	S16	S6	S5	S4	S3
W50	75.6	39.2	78.3	40.9	79.2	79	79	81.5	78.6	78.7	79.1
W36	76.9	40.7	80.7	41.5	81.8	81.4	81.4	83	81	81	81.5
W32	36	80.5	44.8	86.7	44.8	44.5	44.5	42.5	44.6	44.4	44.2
W31	66.9	35.3	79.5	40.6	79.5	79.4	79.4	83	79.7	79.3	78.8
W29	69.3	37.5	87.1	43.1	86.7	86.7	86.7	92.7	88.5	86.3	85.8
W25	33.4	75.3	41.9	81	41.9	41.7	41.7	39.7	41.7	41.7	41.5
W24	35.7	81	44.5	86.3	44.6	44.3	44.3	42.1	44.3	44.2	44
W10	35	79.6	43.9	85.1	44	43.8	43.8	41.6	43.8	43.7	43.4
W8	69.8	38.1	72	39.2	72.2	72	72	75.5	72	71.6	72
S43	77.6	41.1	78.5	40.7	79.6	79	79	77.7	79	78.5	79
S42	100	37.7	72.3	36.7	73.1	72.9	72.9	71.9	72	72.2	72.6
S40	37.7	100	41.6	76.7	42.1	41.9	41.9	39.3	41.4	41.4	41.6
S29	72.3	41.6	100	42.9	96.9	96.5	96.5	93	97.5	96.5	95.9
S26	36.7	76.7	42.9	100	42.8	42.8	42.8	44.6	43.2	42.7	42.4
S23	73.1	42.1	96.9	42.8	100	98.2	98.2	91.8	96.3	97.2	97.6
S17	72.9	41.9	96.5	42.8	98.2	100	100	91.7	96.2	97.4	97.4
S16	72.9	41.9	96.5	42.8	98.2	100	100	91.7	96.2	97.4	97.4
S6	71.9	39.3	93	44.6	91.8	91.7	91.7	100	95.3	91.6	91
S5	72	41.4	97.5	43.2	96.3	96.2	96.2	95.3	100	96	95.4
S4	72.2	41.4	96.5	42.7	97.2	97.4	97.4	91.6	96	100	99.40
S3	72.6	41.6	95.9	42.4	97.6	97.4	97.4	91	95.4	99.40	100
06-02-510	70.6	41	95.5	43.3	95.4	95.4	95.4	91.1	95.7	95	94.6
06-02-508	71	41.4	96.3	42.8	95.6	95.6	95.6	92	96.6	95.5	95
06-02-507	69.5	41.4	94.1	43.5	93.1	93.2	93.2	90	94.4	93.1	92.5
06-02-415	70.9	41.4	94.8	43.5	95.3	95.1	95.1	89.9	94.5	94.6	94.1
06-02-409	68.4	40.1	91	42.1	91	91.5	91.5	86.6	91.2	91	90.5
06-02-406	68.9	41	93	42.9	92.9	92.9	92.9	89.1	93.6	92.6	92.1
06-02-405	71	41.5	94.9	43.4	95.4	95.8	95.8	90.7	95.3	95.3	94.9
06-02-401	69.6	41.1	93.5	42.9	93.5	93.7	93.7	89.8	94.4	93.6	93
06-02-310	67.8	40.1	91.7	41.8	91.7	91.7	91.7	87.6	92.3	91.8	91.4
06-02-309	68	40.2	91.9	41.8	91.9	91.9	91.9	88	92.6	92	91.6
06-02-306	69.7	40.9	93.8	42.9	94.1	94.1	94.1	89.8	94.4	93.6	93.1
06-02-304	69.7	41.4	94.1	43.7	93.3	93.4	93.4	89.5	94.1	93	92.5
06-02-303	69.1	41.3	94	43.3	93.3	93.3	93.3	89.8	94.4	93	92.5
06-02-210	69.6	41.1	94	43.2	94.3	94.4	94.4	89.9	94.5	93.8	93.3
06-02-207	65	38.2	84.4	41	84.3	84.9	84.9	80.9	85.3	84.8	84.3
06-02-110	70.7	41.2	94.4	43	95	95	95	90.2	94.7	94.9	94.5
06-02-108	71.9	41.3	95.5	43.1	96.2	96.7	96.7	91.1	95.3	96.9	96.5
06-02-104	70.9	41.4	95.5	43.4	95.9	95.7	95.7	90.9	95.4	95.3	94.8
06-02-103	71.4	41.6	95.8	43.3	95.5	95.6	95.6	91.3	95.9	94.9	94.4
02-02-508	71.6	41.4	95.8	43.1	95.2	95.1	95.1	91.2	95.6	95.1	94.6
02-02-503	70	40.9	94.5	43	94.3	94.3	94.3	90.5	95	93.9	93.4
02-02-501	70.7	41.3	95.5	43.1	95.1	95.3	95.3	91.1	95.4	94.5	94.1
02-02-418	70.9	41.5	95.5	43	95.2	95.3	95.3	91.1	95.7	95	94.5
02-02-329	68.5	39.8	91.5	41.1	91.7	91.3	91.3	87.3	92	91.1	90.6
02-02-309	71.2	41.3	95.9	43.6	96.5	96.4	96.4	91.4	95.9	95.8	95.3
02-02-306	69.8	41	93.5	43.5	93	92.9	92.9	89.1	93.6	92.7	92.2
02-02-304	67	40	90.7	41.4	90.5	90.9	90.9	86.7	91.4	90.2	89.7
02-02-223	69.2	41	93.4	43.1	92.7	92.8	92.8	88.9	93.5	92.4	91.9

	06-02-510	06-02-508	06-02-507	06-02-415	06-02-409	06-02-406
W50	77.1	77.7	76	77	75.2	75.7
W36	79.8	80.1	78	79.6	77	77.7
W32	44.2	44.4	44.5	44.7	42.9	44.1
W31	79.3	79.6	78.1	78.7	75.9	77.6
W29	*87.3*	86.3	86.4	85.9	84.5	85.8
W25	42.1	41.5	42.8	42.7	41	42.4
W24	44.1	44.2	44.4	44.5	42.8	44
W10	43.5	43.6	43.8	44.1	42.2	43.4
W8	71.6	71.2	70.7	71.3	68.9	70
S43	78.6	78	79.4	78.4	78.1	79.6
S42	70.6	71	69.5	70.9	68.4	68.9
S40	41	41.4	41.4	41.4	40.1	41
S29	*95.5*	*96.3*	*94.1*	*94.8*	*91*	*93*
S26	43.3	42.8	43.5	43.5	42.1	42.9
S23	*95.4*	*95.6*	*93.1*	*95.3*	*91*	*92.9*
S17	*95.4*	*95.6*	*93.2*	*95.1*	*91.5*	*92.9*
S16	*95.4*	*95.6*	*93.2*	*95.1*	*91.5*	*92.9*
S6	*91.1*	*92*	*90*	*89.9*	86.6	*89.1*
S5	*95.7*	*96.6*	*94.4*	*94.5*	*91.2*	*93.6*
S4	*95*	*95.5*	*93.1*	*94.6*	*91*	*92.6*
S3	*94.6*	*95*	*92.5*	*94.1*	*90.5*	*92.1*
06-02-510	100	*96.6*	*94.6*	*96.1*	*92.2*	*94*
06-02-508	*96.6*	100	*95*	*95.5*	*92*	*94.3*
06-02-507	*94.6*	*95*	100	*94*	*93.6*	*96.5*
06-02-415	*96.1*	*95.5*	*94*	100	*91.8*	*94*
06-02-409	*92.2*	*92*	*93.6*	*91.8*	100	*92.6*
06-02-406	*94*	*94.3*	*96.5*	*94*	*92.6*	100
06-02-405	*95.7*	*95.9*	*94.1*	*95.5*	*91.4*	*94.5*
06-02-401	*94.4*	*95*	*94.4*	*93.7*	*92*	*94.1*
06-02-310	*92.7*	*92.9*	*92.9*	*92.3*	*90.1*	*93.3*
06-02-309	*93.3*	*93.6*	*93.6*	*92.3*	*90.7*	*93.2*
06-02-306	*95*	*94.8*	*95.8*	*95*	*92.6*	*96.6*
06-02-304	*94.4*	*94.5*	*97.3*	*94.5*	*93.3*	*96.7*
06-02-303	*94.4*	*95.4*	*95.9*	*94*	*92.7*	*95.6*
06-02-210	*95.4*	*95*	*96.3*	*95.6*	*93.3*	*97*
06-02-207	86.4	85.6	*87*	85.2	86.1	86.9
06-02-110	*95.4*	*95.5*	*93.3*	*95*	*90.8*	*93.4*
06-02-108	*95.4*	*95.4*	*92.8*	*94.7*	*90.9*	*92.9*
06-02-104	*96.7*	*96.3*	*94.7*	*96.8*	*92.1*	*94.3*
06-02-103	*95.9*	*96.5*	*95.1*	*95.8*	*91.8*	*94.9*
02-02-508	*95.8*	*96.6*	*95.4*	*95.2*	*91.3*	*94.4*
02-02-503	*95.9*	*95.1*	*94.6*	*95.4*	*91.8*	*94.2*
02-02-501	*96.1*	*95.9*	*93.8*	*94.8*	*91.2*	*93.2*
02-02-418	*96.7*	*96.5*	*94.3*	*95.1*	*91.6*	*93.5*
02-02-329	*92.7*	*92.7*	*93.2*	*91.3*	*90.9*	*92.8*
02-02-309	*97.3*	*96.8*	*95.2*	*97.5*	*92.7*	*94.9*
02-02-306	*94.2*	*93.7*	*96.7*	*94.1*	*92.4*	*96.3*
02-02-304	*91.2*	*91.8*	*92.2*	*90.7*	*89.2*	*92.1*
02-02-223	*94.1*	*93.9*	*97.4*	*93.8*	*93*	*97.2*



	06-02-405	06-02-401	06-02-309	06-02-306	06-02-304	06-02-303
W50	77.7	76.1	74.7	75.7	76	75.4
W36	80.2	78.7	77.3	78.4	78.2	77.6
W32	45.1	44.5	43.4	44	44.8	44.4
W31	79.6	78.5	76.8	78.1	77.8	77.4
W29	86	86.3	84.8	86.6	85.9	86.1
W25	42.6	41.9	41.2	42.5	42.7	42.3
W24	44.9	44.5	43.3	44.1	44.7	44.3
W10	44.4	43.7	42.8	43.4	44.1	43.7
W8	70.9	69.5	68	70	70.6	70.2
S43	78	77.2	76.9	79.2	79.2	78.4
S42	71	69.6	68	69.7	69.7	69.1
S40	41.5	41.1	40.2	40.9	41.4	41.3
S29	*94.9*	*93.5*	*91.9*	*93.8*	*94.1*	*94*
S26	43.4	42.9	41.8	42.9	43.7	43.3
S23	*95.4*	*93.5*	*91.9*	*94.1*	*93.3*	*93.3*
S17	*95.8*	*93.7*	*91.9*	*94.1*	*93.4*	*93.3*
S16	*95.8*	*93.7*	*91.9*	*94.1*	*93.4*	*93.3*
S6	*90.7*	*89.8*	*88*	*89.8*	*89.5*	*89.8*
S5	*95.3*	*94.4*	*92.6*	*94.4*	*94.1*	*94.4*
S4	*95.3*	*93.6*	*92*	*93.6*	*93*	*93*
S3	*94.9*	*93*	*91.6*	*93.1*	*92.5*	*92.5*
06-02-510	*95.7*	*94.4*	*93.3*	*95*	*94.4*	*94.4*
06-02-508	*95.9*	*95*	*93.6*	*94.8*	*94.5*	*95.4*
06-02-507	*94.1*	*94.4*	*93.6*	*95.8*	*97.3*	*95.9*
06-02-415	*95.5*	*93.7*	*92.3*	*95*	*94.5*	*94*
06-02-409	*91.4*	*92*	*90.7*	*92.6*	*93.3*	*92.7*
06-02-406	*94.5*	*94.1*	*93.2*	*96.6*	*96.7*	*95.6*
06-02-405	100	*94.9*	*92.7*	*95.2*	*94.5*	*94.1*
06-02-401	*94.9*	100	*93.4*	*94.5*	*94*	*94.1*
06-02-310	*92.7*	*92.8*	99.1	*93.1*	*93.1*	*92.9*
06-02-309	*92.7*	*93.4*	100	*93.3*	*93*	*93.2*
06-02-306	*95.2*	*94.5*	*93.3*	100	*95.5*	*95.7*
06-02-304	*94.5*	*94*	*93*	*95.5*	100	*95.9*
06-02-303	*94.1*	*94.1*	*93.2*	*95.7*	*95.9*	100
06-02-210	*95.5*	*95*	*94.1*	*98.1*	*96.1*	*96.2*
06-02-207	86	86	84.5	*87.5*	86.3	86.3
06-02-110	*96.6*	*95.8*	*92.5*	*94.3*	*93.4*	*93.2*
06-02-108	*95.6*	*93.8*	*92*	*93.9*	*92.8*	*92.9*
06-02-104	*95.9*	*94*	*92.6*	*95.7*	*94.8*	*95*
06-02-103	*96.2*	*94.1*	*92.7*	*94.5*	*95.7*	*94.2*
02-02-508	*95.7*	*94*	*92.4*	*94*	*95.2*	*93.9*
02-02-503	*95.1*	*94.7*	*93.2*	*94.8*	*94.6*	*94.2*
02-02-501	*95.5*	*93.8*	*92*	*94*	*93.8*	*93.4*
02-02-418	*96*	*95*	*93.2*	*94.1*	*93.9*	*93.6*
02-02-329	*91.9*	*93.2*	*95.6*	*93*	*92.7*	*93*
02-02-309	*96.5*	*94.7*	*93.1*	*96.4*	*95.1*	*95.3*
02-02-306	*94.1*	*93.1*	*93.1*	*96*	*96.2*	*94.8*
02-02-304	*91.8*	*91.8*	*93.6*	*92.1*	*92.4*	*92.3*
02-02-223	*94.1*	*93.8*	*93.3*	*96.5*	*96.8*	*94.9*

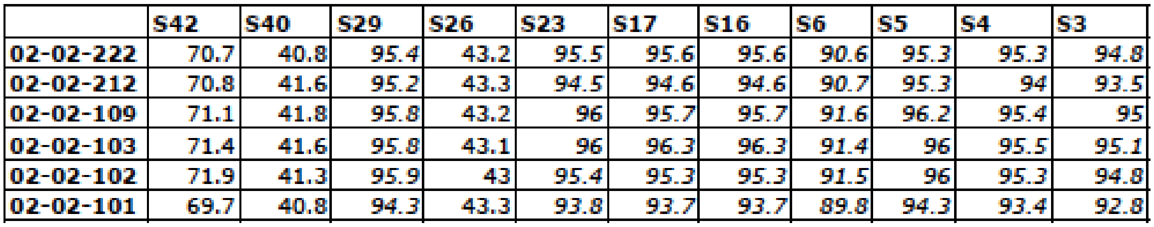
	06-02-210	06-02-110	06-02-108	06-02-104	06-02-103	02-02-508
W50	76.1	77.2	78.7	77.3	78.1	78.2
W36	78.8	79.6	81	79.8	80.1	80.1
W32	44.4	44.8	44.8	44.5	45	44.7
W31	78.2	79.6	80	79.4	80	79.7
W29	87	85.6	86.5	86.3	86.3	85.9
W25	42.9	42.3	41.8	42.5	42.3	42
W24	44.3	44.6	44.6	44.4	44.8	44.5
W10	43.7	44.1	44	43.8	44.2	44.1
W8	70.3	70.2	71.1	71.2	70.7	71.1
S43	79.9	77.6	78	78.5	78.1	77.9
S42	69.6	70.7	71.9	70.9	71.4	71.6
S40	41.1	41.2	41.3	41.4	41.6	41.4
S29	94	94.4	95.5	95.5	95.8	95.8
S26	43.2	43	43.1	43.4	43.3	43.1
S23	94.3	95	96.2	95.9	95.5	95.2
S17	94.4	95	96.7	95.7	95.6	95.1
S16	94.4	95	96.7	95.7	95.6	95.1
S6	89.9	90.2	91.1	90.9	91.3	91.2
S5	94.5	94.7	95.3	95.4	95.9	95.6
S4	93.8	94.9	96.9	95.3	94.9	95.1
S3	93.3	94.5	96.5	94.8	94.4	94.6
06-02-510	95.4	95.4	95.4	96.7	95.9	95.8
06-02-508	95	95.5	95.4	96.3	96.5	96.6
06-02-507	96.3	93.3	92.8	94.7	95.1	95.4
06-02-415	95.6	95	94.7	96.8	95.8	95.2
06-02-409	93.3	90.8	90.9	92.1	91.8	91.3
06-02-406	97	93.4	92.9	94.3	94.9	94.4
06-02-405	95.5	96.6	95.6	95.9	96.2	95.7
06-02-401	95	95.8	93.8	94	94.1	94
06-02-310	94.1	92.4	91.8	92.4	92.7	92.3
06-02-309	94.1	92.5	92	92.6	92.7	92.4
06-02-306	98.1	94.3	93.9	95.7	94.5	94
06-02-304	96.1	93.4	92.8	94.8	95.7	95.2
06-02-303	96.2	93.2	92.9	95	94.2	93.9
06-02-210	100	94.6	94.2	96.1	95.2	94.5
06-02-207	88	85.8	85	85.6	85.5	85.2
06-02-110	94.6	100	95.1	95.9	95.8	95.3
06-02-108	94.2	95.1	100	95.3	95.1	95.3
06-02-104	96.1	95.9	95.3	100	96.4	95.8
06-02-103	95.2	95.8	95.1	96.4	100	97.3
02-02-508	94.5	95.3	95.3	95.8	97.3	100
02-02-503	95.6	94.7	94	95.4	95.6	94.9
02-02-501	94.3	95.1	94.9	95.5	96.2	95.8
02-02-418	94.5	95.5	95.3	95.7	96.4	96.3
02-02-329	93.3	91.4	91.1	92.4	92.6	92.3
02-02-309	96.6	96	96	98	96.8	96.2
02-02-306	96.6	93.6	92.6	94.4	94.6	94.6
02-02-304	93	90.9	90.5	91.4	92.4	91.7
02-02-223	96.8	93.4	92.6	94.4	95.1	95.1



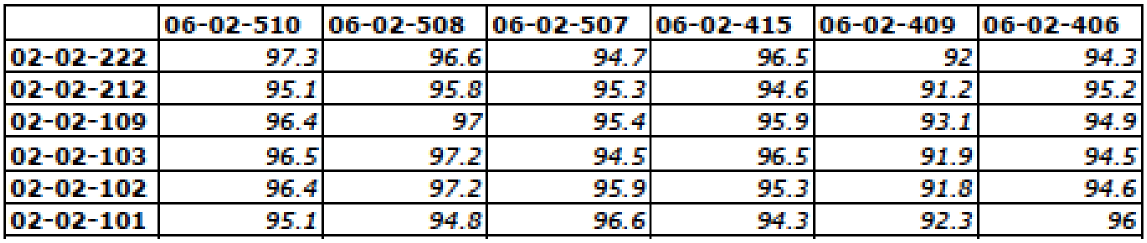
	02-02-501	**02-02-418**	**02-02-329**	**02-02-309**	**02-02-306**	**02-02-304**
W50	77.3	77.9	74.1	77.7	75.6	75
W36	79.8	80.5	76.8	80.4	77.7	77.8
W32	44.7	44.7	42.9	44.7	44.4	43.6
W31	79.2	80	76.6	79.9	77.4	77.6
W29	86.3	86.4	84.4	*87.1*	85.8	83.7
W25	41.9	42	40.7	42.6	43	41
W24	44.5	44.5	43	44.5	44.3	43.5
W10	43.9	43.9	42.4	43.9	43.8	42.8
W8	71.2	71	67.5	71.9	70.3	67.9
S43	77.5	78	76.8	78.7	80.3	75.3
S42	70.7	70.9	68.5	71.2	69.8	68.5
S40	41.3	41.5	39.8	41.3	41	39.7
S29	*95.5*	*95.5*	*91.5*	*95.9*	*93.5*	*90.8*
S26	43.1	43	41.1	43.6	43.5	40.4
S23	*95.1*	*95.2*	*91.7*	*96.5*	*93*	*90.6*
S17	*95.3*	*95.3*	*91.3*	*96.4*	*92.9*	*91*
S16	*95.3*	*95.3*	*91.3*	*96.4*	*92.9*	*91*
S6	*91.1*	*91.1*	*87.3*	*91.4*	*89.1*	86.8
S5	*95.4*	*95.7*	*92*	*95.9*	*93.6*	*91.4*
S4	*94.5*	*95*	*91.1*	*95.8*	*92.7*	*90.3*
S3	*94.1*	*94.5*	*90.6*	*95.3*	*92.2*	*90*
06-02-510	*96.1*	*96.7*	*92.7*	*97.3*	*94.2*	*91.3*
06-02-508	*95.9*	*96.5*	*92.7*	*96.8*	*93.7*	*91.9*
06-02-507	*93.8*	*94.3*	*93.2*	*95.2*	*96.7*	*92.3*
06-02-415	*94.8*	*95.1*	*91.3*	*97.5*	*94.1*	*90.8*
06-02-409	*91.2*	*91.6*	*90.9*	*92.7*	*92.4*	*88.3*
06-02-406	*93.2*	*93.5*	*92.8*	*94.9*	*96.3*	*92.4*
06-02-405	*95.5*	*96*	*91.9*	*96.5*	*94.1*	*91.9*
06-02-401	*93.8*	*95*	*93.2*	*94.7*	*93.1*	*92*
06-02-310	*91.8*	*92.6*	*94.8*	*93*	*93.2*	*93.5*
06-02-309	*92*	*93.2*	*95.6*	*93.1*	*93.1*	*93.6*
06-02-306	*94*	*94.1*	*93*	*96.4*	*96*	*92.2*
06-02-304	*93.8*	*93.9*	*92.7*	*95.1*	*96.2*	*92.5*
06-02-303	*93.4*	*93.6*	*93*	*95.3*	*94.8*	*92.3*
06-02-210	*94.3*	*94.5*	*93.3*	*96.6*	*96.6*	*93.1*
06-02-207	85.3	85	84.6	86.2	86.9	85.1
06-02-110	*95.1*	*95.5*	*91.4*	*96*	*93.6*	*91*
06-02-108	*94.9*	*95.3*	*91.1*	*96*	*92.6*	*91.4*
06-02-104	*95.5*	*95.7*	*92.4*	*98*	*94.4*	*91.4*
06-02-103	*96.2*	*96.4*	*92.6*	*96.8*	*94.6*	*92.5*
02-02-508	*95.8*	*96.3*	*92.3*	*96.2*	*94.6*	*91.7*
02-02-503	*94.9*	*95.8*	*92.7*	*95.9*	*94.1*	*91.9*
02-02-501	100	*96*	*91.6*	*96*	*93.4*	*91.1*
02-02-418	*96*	100	*92.4*	*96.2*	*93.7*	*91.2*
02-02-329	*91.6*	*92.4*	100	*92.6*	*92.5*	*94.2*
02-02-309	*96*	*96.2*	*92.6*	100	*95.1*	*92*
02-02-306	*93.4*	*93.7*	*92.5*	*95.1*	100	*91.4*
02-02-304	*91*	*91.1*	*94.2*	*91.9*	*91.3*	100
02-02-223	*93.6*	*93.9*	*92.8*	*94.7*	*96.8*	*91.3*

	02-02-223	02-02-222	02-02-212	02-02-109	02-02-103	02-02-102	02-02-101
W50	75.6	77.6	77.1	77.4	78.3	78.5	76.3
W36	77.5	80.1	79.1	80.2	80.8	80.4	78.3
W32	44.1	44.2	45.1	44.8	44.9	44.5	44.2
W31	77.6	79.9	78.8	79.5	80.3	80	78.4
W29	85.7	86.4	85.7	*87.1*	*87.1*	86.2	86
W25	42.8	42.3	42.5	42.1	42.2	41.9	42.5
W24	44.1	44.2	44.9	44.7	44.7	44.3	44.1
W10	43.5	43.5	44.2	44.1	44.3	43.9	43.6
W8	69.9	71.5	70.6	70.9	71.1	71.4	70.6
S43	79.5	78.3	77.9	78.6	78.2	78.2	79
S42	69.2	70.7	70.8	71.1	71.4	71.9	69.7
S40	41	40.8	41.6	41.8	41.6	41.3	40.8
S29	*93.4*	*95.4*	*95.2*	*95.8*	*95.8*	*95.9*	*94.3*
S26	43.1	43.2	43.3	43.2	43.1	43	43.3
S23	*92.7*	*95.5*	*94.5*	*96*	*96*	*95.4*	*93.8*
S17	*92.8*	*95.6*	*94.6*	*95.7*	*96.3*	*95.3*	*93.7*
S16	*92.8*	*95.6*	*94.6*	*95.7*	*96.3*	*95.3*	*93.7*
S6	*88.9*	*90.6*	*90.7*	*91.6*	*91.4*	*91.5*	*89.8*
S5	*93.5*	*95.3*	*95.3*	*96.2*	*96*	*96*	*94.3*
S4	*92.4*	*95.3*	*94*	*95.4*	*95.5*	*95.3*	*93.4*
S3	*91.9*	*94.8*	*93.5*	*95*	*95.1*	*94.8*	*92.8*
06-02-510	*94.1*	*97.3*	*95.1*	*96.4*	*96.5*	*96.4*	*95.1*
06-02-508	*93.9*	*96.6*	*95.8*	*97*	*97.2*	*97.2*	*94.8*
06-02-507	*97.4*	*94.7*	*95.3*	*95.4*	*94.5*	*95.9*	*96.6*
06-02-415	*93.8*	*96.5*	*94.6*	*95.9*	*96.5*	*95.3*	*94.3*
06-02-409	*93*	*92*	*91.2*	*93.1*	*91.9*	*91.8*	*92.3*
06-02-406	*97.2*	*94.3*	*95.2*	*94.9*	*94.5*	*94.6*	*96*
06-02-405	*94.1*	*95.9*	*96.5*	*96.2*	*97.1*	*95.9*	*95.1*
06-02-401	*93.8*	*94.2*	*93.7*	*94.9*	*95*	*94.5*	*94.4*
06-02-310	*92.8*	*93*	*92.2*	*93.9*	*93.8*	*92.4*	*92.4*
06-02-309	*93.3*	*93.4*	*92.1*	*94.8*	*93.8*	*92.9*	*92.5*
06-02-306	*96.5*	*95.2*	*94.4*	*95.9*	*95.2*	*94.1*	*95.4*
06-02-304	*96.8*	*94.6*	*95.8*	*95.2*	*95*	*95.4*	*96.6*
06-02-303	*94.9*	*94.6*	*93.9*	*95.3*	*94.7*	*94*	*95.4*
06-02-210	*96.8*	*95.4*	*94.9*	*96.4*	*95.7*	*94.6*	*95.8*
06-02-207	86.9	85.5	85.3	86.1	85.7	85.5	86.6
06-02-110	*93.4*	*95.6*	*95*	*95.8*	*96.5*	*95.5*	*94.3*
06-02-108	*92.6*	*95.4*	*94.2*	*95.3*	*95.7*	*95.4*	*93.5*
06-02-104	*94.4*	*96.9*	*95.2*	*96.6*	*96.9*	*96*	*95.1*
06-02-103	*95.1*	*96.5*	*97.3*	*96.4*	*97.6*	*97.4*	*95.6*
02-02-508	*95.1*	*96.1*	*96.9*	*95.8*	*96.7*	99.4	*95.7*
02-02-503	*94.2*	*95.8*	*94.7*	*95.8*	*96.5*	*95*	*94.2*
02-02-501	*93.6*	*95.5*	*95.4*	*95.9*	*96.4*	*96*	*94.4*
02-02-418	*93.9*	*96.8*	*95.5*	*96.2*	*97.2*	*96.9*	*94.5*
02-02-329	*92.8*	*92.5*	*92.3*	*93.9*	*92.5*	*92.7*	*92.7*
02-02-309	*94.7*	*97.4*	*95.5*	*96.9*	*97.3*	*96.4*	*95.6*
02-02-306	*96.8*	*94.4*	*95*	*95*	*94.4*	*94.8*	*95.7*
02-02-304	*92.1*	*91.2*	*92.1*	*92.2*	*92.2*	*91.7*	*92.4*
02-02-223	100	*94.6*	*95.4*	*94.7*	*94.2*	*95.4*	*96.6*

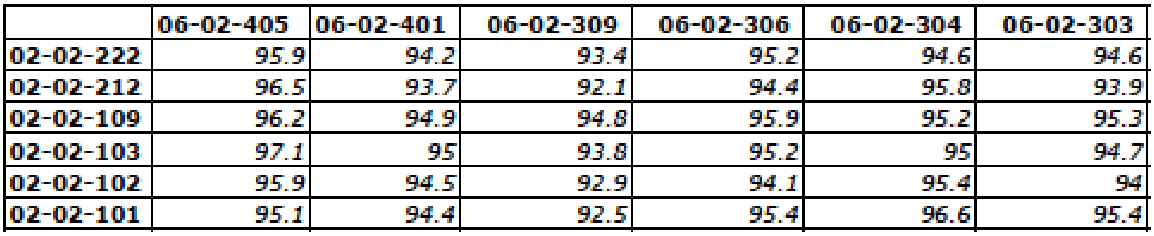
	W50	W36	W32	W31	W29	W25	W24	W10	W8	S43
02-02-222	77.6	80.1	44.2	79.9	86.4	42.3	44.2	43.5	70.9	78.3
02-02-212	77.1	79.1	45.1	78.8	85.7	42.5	44.9	44.2	71.5	77.9
02-02-109	77.4	80.2	44.8	79.5	*87.1*	42.1	44.7	44.1	72	78.6
02-02-103	78.3	80.8	44.9	80.3	*87.1*	42.2	44.7	44.3	72	78.2
02-02-102	78.5	80.4	44.5	80	86.2	41.9	44.3	43.9	72.4	78.2
02-02-101	76.3	78.3	44.2	78.4	86	42.5	44.1	43.6	71.6	79



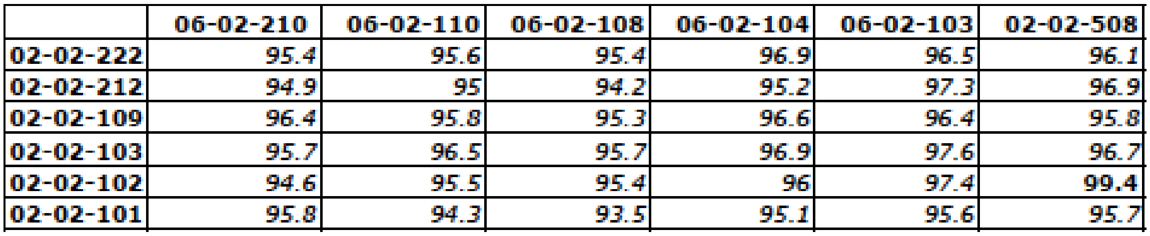
	S42	S40	S29	S26	S23	S17	S16	S6	S5	S4	S3
02-02-222	70.7	40.8	*95.4*	43.2	*95.5*	*95.6*	*95.6*	*90.6*	*95.3*	*95.3*	*94.8*
02-02-212	70.8	41.6	*95.2*	43.3	*94.5*	*94.6*	*94.6*	*90.7*	*95.3*	*94*	*93.5*
02-02-109	71.1	41.8	*95.8*	43.2	*96*	*95.7*	*95.7*	*91.6*	*96.2*	*95.4*	*95*
02-02-103	71.4	41.6	*95.8*	43.1	*96*	*96.3*	*96.3*	*91.4*	*96*	*95.5*	*95.1*
02-02-102	71.9	41.3	*95.9*	43	*95.4*	*95.3*	*95.3*	*91.5*	*96*	*95.3*	*94.8*
02-02-101	69.7	40.8	*94.3*	43.3	*93.8*	*93.7*	*93.7*	*89.8*	*94.3*	*93.4*	*92.8*



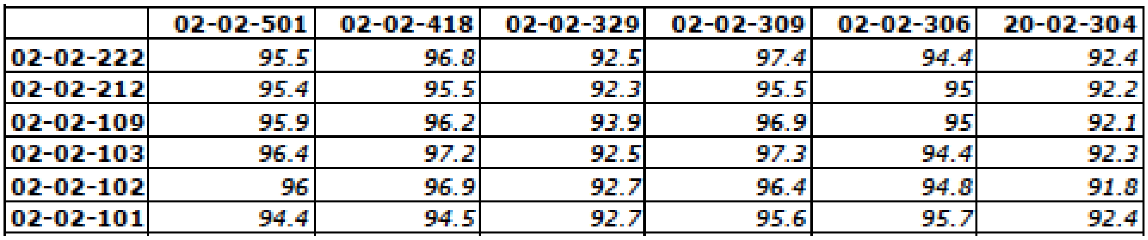
	06-02-510	06-02-508	06-02-507	06-02-415	06-02-409	06-02-406
02-02-222	*97.3*	*96.6*	*94.7*	*96.5*	*92*	*94.3*
02-02-212	*95.1*	*95.8*	*95.3*	*94.6*	*91.2*	*95.2*
02-02-109	*96.4*	*97*	*95.4*	*95.9*	*93.1*	*94.9*
02-02-103	*96.5*	*97.2*	*94.5*	*96.5*	*91.9*	*94.5*
02-02-102	*96.4*	*97.2*	*95.9*	*95.3*	*91.8*	*94.6*
02-02-101	*95.1*	*94.8*	*96.6*	*94.3*	*92.3*	*96*



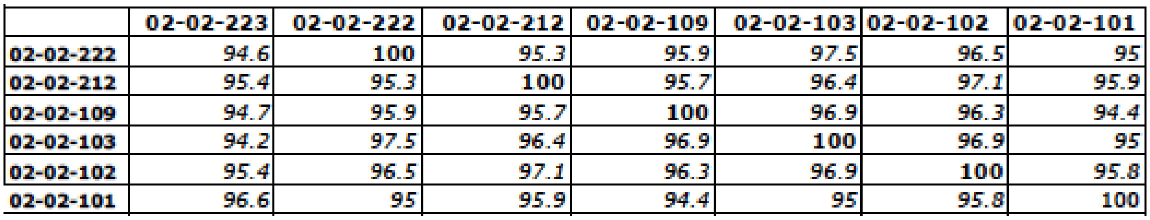
	06-02-405	06-02-401	06-02-309	06-02-306	06-02-304	06-02-303
02-02-222	*95.9*	*94.2*	*93.4*	*95.2*	*94.6*	*94.6*
02-02-212	*96.5*	*93.7*	*92.1*	*94.4*	*95.8*	*93.9*
02-02-109	*96.2*	*94.9*	*94.8*	*95.9*	*95.2*	*95.3*
02-02-103	*97.1*	*95*	*93.8*	*95.2*	*95*	*94.7*
02-02-102	*95.9*	*94.5*	*92.9*	*94.1*	*95.4*	*94*
02-02-101	*95.1*	*94.4*	*92.5*	*95.4*	*96.6*	*95.4*



	06-02-210	06-02-110	06-02-108	06-02-104	06-02-103	02-02-508
02-02-222	*95.4*	*95.6*	*95.4*	*96.9*	*96.5*	*96.1*
02-02-212	*94.9*	*95*	*94.2*	*95.2*	*97.3*	*96.9*
02-02-109	*96.4*	*95.8*	*95.3*	*96.6*	*96.4*	*95.8*
02-02-103	*95.7*	*96.5*	*95.7*	*96.9*	*97.6*	*96.7*
02-02-102	*94.6*	*95.5*	*95.4*	*96*	*97.4*	99.4
02-02-101	*95.8*	*94.3*	*93.5*	*95.1*	*95.6*	*95.7*



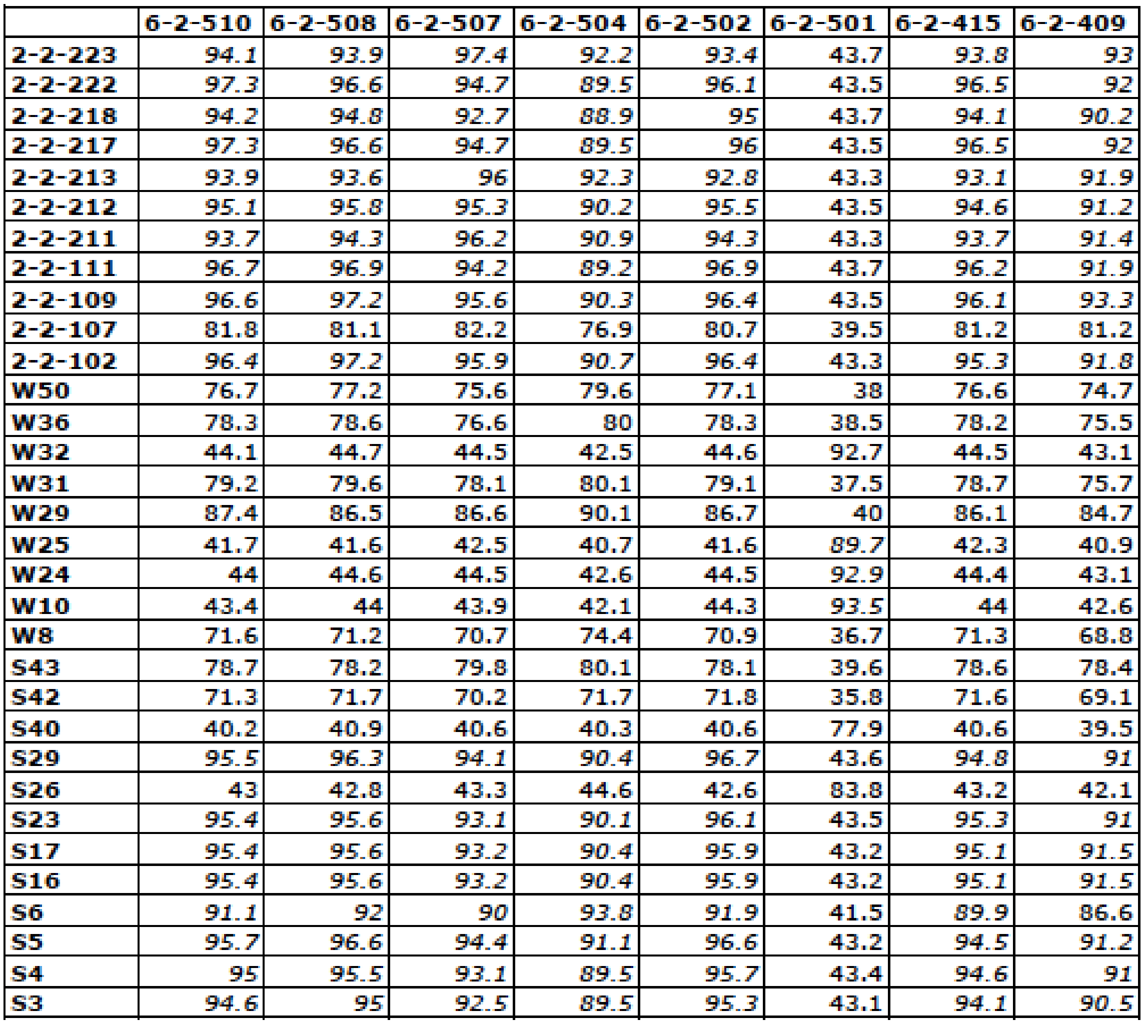
	02-02-501	02-02-418	02-02-329	02-02-309	02-02-306	20-02-304
02-02-222	*95.5*	*96.8*	*92.5*	*97.4*	*94.4*	*92.4*
02-02-212	*95.4*	*95.5*	*92.3*	*95.5*	*95*	*92.2*
02-02-109	*95.9*	*96.2*	*93.9*	*96.9*	*95*	*92.1*
02-02-103	*96.4*	*97.2*	*92.5*	*97.3*	*94.4*	*92.3*
02-02-102	*96*	*96.9*	*92.7*	*96.4*	*94.8*	*91.8*
02-02-101	*94.4*	*94.5*	*92.7*	*95.6*	*95.7*	*92.4*



	02-02-223	02-02-222	02-02-212	02-02-109	02-02-103	02-02-102	02-02-101
02-02-222	*94.6*	100	*95.3*	*95.9*	*97.5*	*96.5*	*95*
02-02-212	*95.4*	*95.3*	100	*95.7*	*96.4*	*97.1*	*95.9*
02-02-109	*94.7*	*95.9*	*95.7*	100	*96.9*	*96.3*	*94.4*
02-02-103	*94.2*	*97.5*	*96.4*	*96.9*	100	*96.9*	*95*
02-02-102	*95.4*	*96.5*	*97.1*	*96.3*	*96.9*	100	*95.8*
02-02-101	*96.6*	*95*	*95.9*	*94.4*	*95*	*95.8*	100

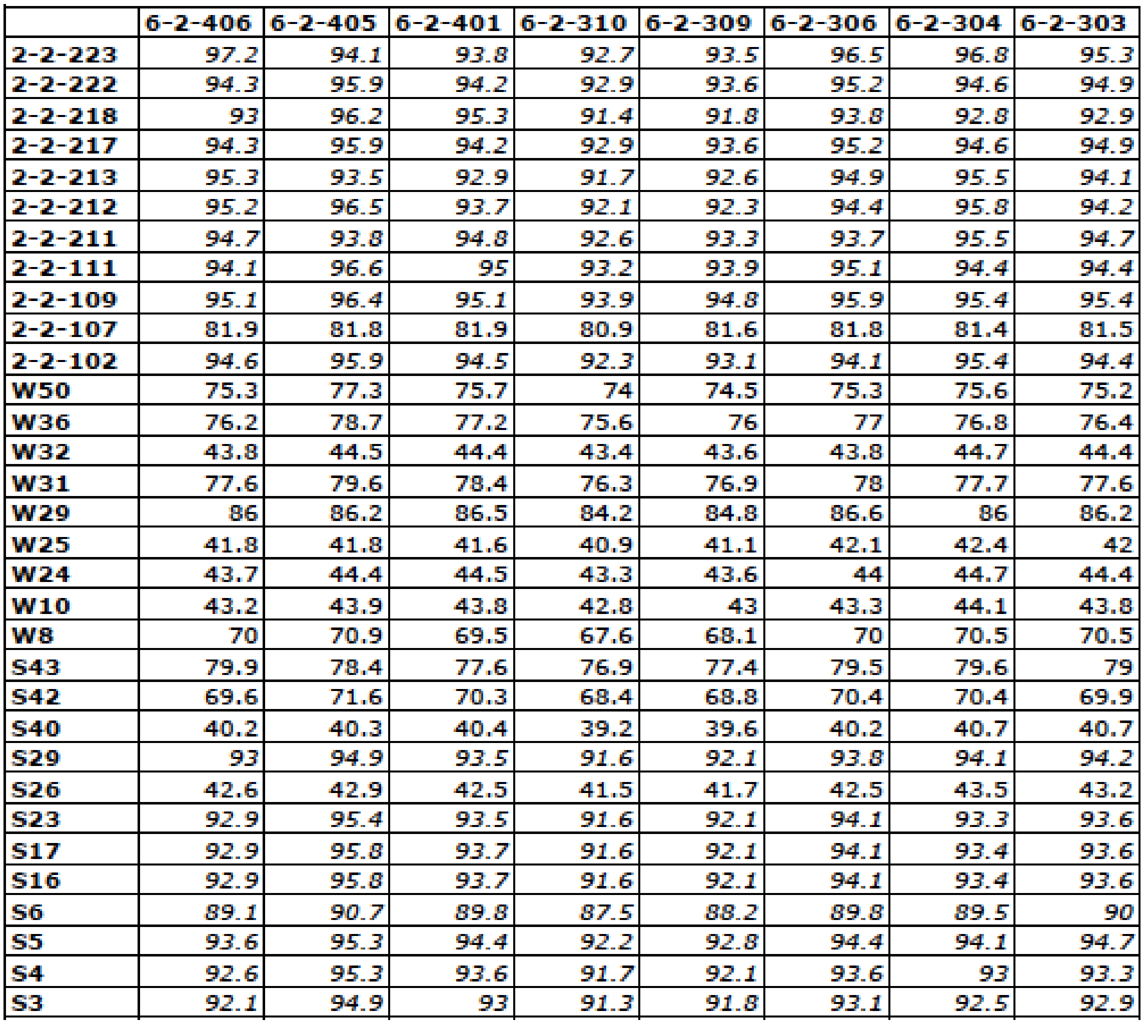
Table A3: Complete similarity matrix for summer and winter clones that were most closely related to *Methanosaeta concilii* compared to Chen's clones that were most closely related to *Methanosaeta concilii*. Italicized numbers indicate a similarity index of 87 or greater; bolded numbers indicate sequences with a similarity index of 98.7 or greater.

	6-2-510	6-2-508	6-2-507	6-2-504	6-2-502	6-2-501	6-2-415	6-2-409
6-2-510	**100**	*96.6*	*94.6*	*89.3*	*96*	43.1	*96.1*	*92.2*
6-2-508	*96.6*	**100**	*95*	*89.7*	*97.4*	43.1	*95.5*	*92*
6-2-507	*94.6*	*95*	**100**	*93.7*	*94*	43.7	*94*	*93.6*
6-2-504	*89.3*	*89.7*	*93.7*	**100**	*89.2*	42.1	*89*	*88.3*
6-2-502	*96*	*97.4*	*94*	*89.2*	**100**	43.2	*95.1*	*91.1*
6-2-501	43.1	43.1	43.7	42.1	43.2	**100**	43.7	42.6
6-2-415	*96.1*	*95.5*	*94*	*89*	*95.1*	43.7	**100**	*91.8*
6-2-409	*92.2*	*92*	*93.6*	*88.3*	*91.1*	42.6	*91.8*	**100**
6-2-406	*94*	*94.3*	*96.5*	*91.4*	*93.5*	43.3	*94*	*92.6*
6-2-405	*95.7*	*95.9*	*94.1*	*89*	*95.6*	43.3	*95.5*	*91.4*
6-2-401	*94.4*	*95*	*94.4*	*89*	*93.9*	43	*93.7*	*92*
6-2-310	*92.6*	*92.8*	*92.8*	*87.9*	*92.4*	42.3	*92.2*	*90*
6-2-309	*93.5*	*93.7*	*93.7*	*88.6*	*92.8*	42.6	*92.5*	*90.9*
6-2-306	*95*	*94.8*	*95.8*	*90.5*	*94.3*	43.3	*95*	*92.6*
6-2-304	*94.4*	*94.5*	*97.3*	*92.3*	*93.9*	43.9	*94.5*	*93.3*
6-2-303	*94.7*	*95.6*	*96.3*	*91*	*95.3*	43.5	*94.1*	*93*
6-2-210	*95.4*	*95*	*96.3*	*91.1*	*94.5*	43.6	*95.6*	*93.3*
6-2-207	86.8	86	*87.3*	82.5	85.4	41	85.5	86.3
6-2-206	*95.1*	*95.4*	*95.3*	*90.4*	*95.2*	43	*94.8*	*91.2*
6-2-203	*93.4*	*93.5*	*96.6*	*91.7*	*93.2*	43.5	*93.2*	*91.8*
6-2-120	*95.9*	*95.7*	*96.1*	*90.7*	*95.2*	43.5	*95.4*	*91.9*
6-2-119	*95.2*	*95.7*	*93.7*	*89.1*	*94.8*	43.2	*94.4*	*91.3*
6-2-118	*96.8*	*98.1*	*94.8*	*89.6*	*97.5*	43.3	*95.8*	*91.8*
6-2-117	*95.7*	*97.1*	*93.8*	*88.4*	*97.1*	43.5	*95*	*91*
6-2-113	*95.3*	*95.8*	*93.8*	*89*	*94.9*	43.3	*94.5*	*91.4*
6-2-112	*95.6*	*96.9*	*93.6*	*90.4*	*97.7*	43.6	*94.8*	*90.6*
6-2-111	*95.6*	*96.9*	*93.6*	*90.4*	*97.7*	43.6	*94.8*	*90.6*
6-2-110	*95.4*	*95.5*	*93.3*	*88.5*	*95.4*	43.8	*95*	*90.8*
6-2-108	*95.4*	*95.4*	*92.8*	*88.9*	*95.5*	43.1	*94.7*	*90.9*
6-2-106	*95.6*	*95.3*	*94.9*	*89.8*	*94.9*	43.8	*95.8*	*92.7*
6-2-105	*95.6*	*95.3*	*94.9*	*89.8*	*94.9*	43.8	*95.8*	*92.7*
6-2-104	*96.7*	*96.3*	*94.7*	*89.6*	*96.1*	43.7	*96.8*	*92.1*
6-2-103	*95.9*	*96.5*	*95.1*	*90.1*	*96.3*	43.5	*95.8*	*91.8*
2-2-508	*95.8*	*96.6*	*95.4*	*90.4*	*96.2*	43.4	*95.2*	*91.3*
2-2-503	*95.9*	*95.1*	*94.6*	*89.6*	*94.6*	42.9	*95.4*	*91.8*
2-2-501	*96.1*	*95.9*	*93.8*	*89.1*	*95.8*	43.3	*94.8*	*91.2*
2-2-418	*96.7*	*96.5*	*94.3*	*89.1*	*95.7*	43.1	*95.1*	*91.6*
2-2-402	*96*	*97*	*94*	*88.7*	*96*	42.7	*95.3*	*91.3*
2-2-330	82.8	82.6	84	80.4	82.8	40.1	82.5	83.1
2-2-329	*92.9*	*92.9*	*93.4*	*88.1*	*92.2*	41.8	*91.5*	*91.1*
2-2-324	78.7	78.9	79.1	75.2	78.2	38	78.1	78.7
2-2-320	*93.2*	*93.4*	*97.1*	*93.2*	*92.5*	42.9	*92.5*	*91.8*
2-2-318	*96.1*	*95.3*	*92.7*	*89.1*	*96*	43.3	*94.5*	*90.4*
2-2-317	*94.3*	*94.7*	*94*	*90.8*	*95.2*	43.1	*93.5*	*90.3*
2-2-309	*97.3*	*96.8*	*95.2*	*90*	*96.5*	43.8	*97.5*	*92.7*
2-2-306	*94.2*	*93.7*	*96.7*	*91.5*	*93.7*	44	*94.1*	*92.4*
2-2-305	*91*	*91.6*	*92*	*88.7*	*92*	42.2	*90.3*	*89*
2-2-304	*91.4*	*92*	*92.4*	*87.3*	*91.7*	42	*90.9*	*89.3*

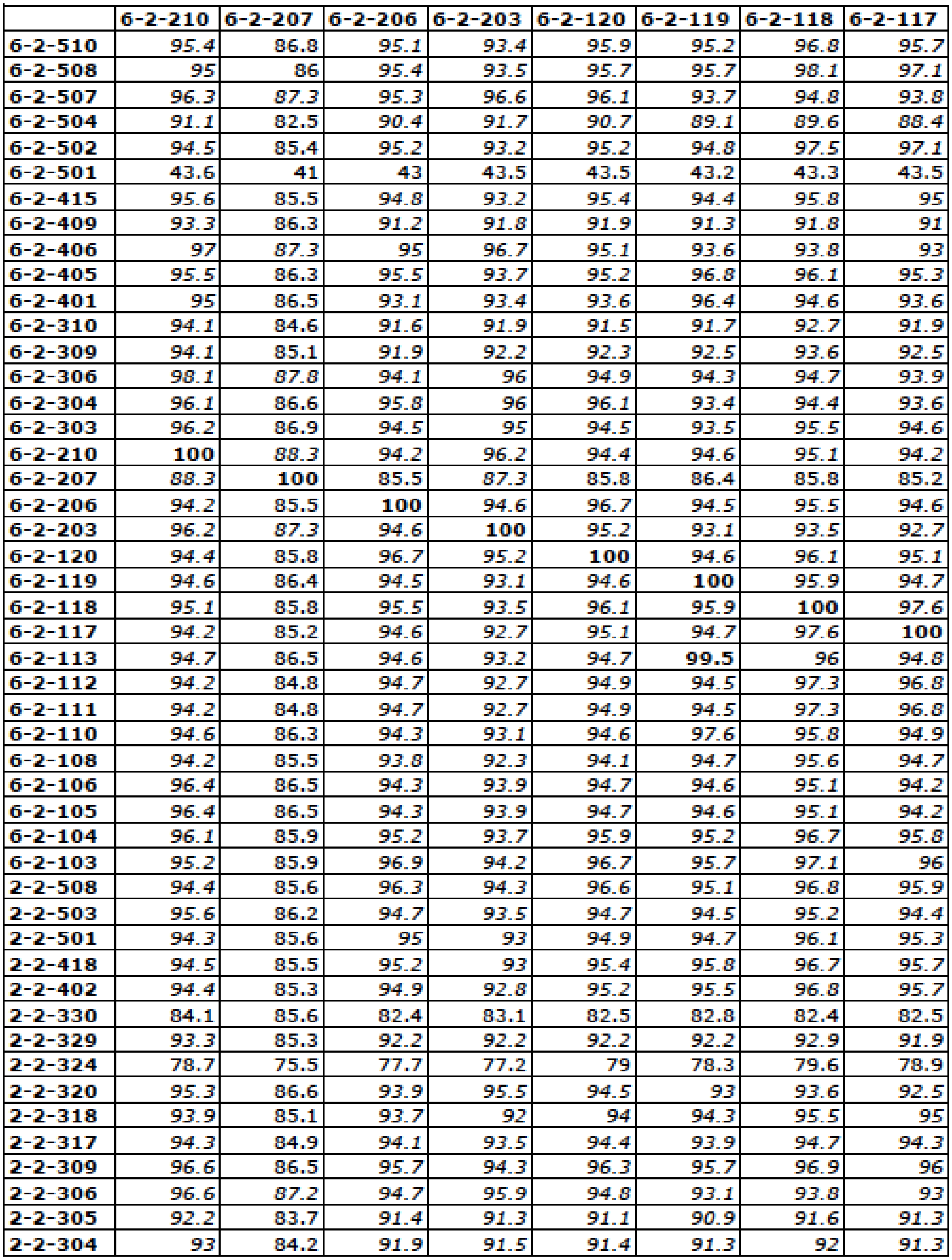


	6-2-510	**6-2-508**	**6-2-507**	**6-2-504**	**6-2-502**	**6-2-501**	**6-2-415**	**6-2-409**
2-2-223	*94.1*	*93.9*	*97.4*	*92.2*	*93.4*	43.7	*93.8*	*93*
2-2-222	*97.3*	*96.6*	*94.7*	*89.5*	*96.1*	43.5	*96.5*	*92*
2-2-218	*94.2*	*94.8*	*92.7*	*88.9*	*95*	43.7	*94.1*	*90.2*
2-2-217	*97.3*	*96.6*	*94.7*	*89.5*	*96*	43.5	*96.5*	*92*
2-2-213	*93.9*	*93.6*	*96*	*92.3*	*92.8*	43.3	*93.1*	*91.9*
2-2-212	*95.1*	*95.8*	*95.3*	*90.2*	*95.5*	43.5	*94.6*	*91.2*
2-2-211	*93.7*	*94.3*	*96.2*	*90.9*	*94.3*	43.3	*93.7*	*91.4*
2-2-111	*96.7*	*96.9*	*94.2*	*89.2*	*96.9*	43.7	*96.2*	*91.9*
2-2-109	*96.6*	*97.2*	*95.6*	*90.3*	*96.4*	43.5	*96.1*	*93.3*
2-2-107	81.8	81.1	82.2	76.9	80.7	39.5	81.2	81.2
2-2-102	*96.4*	*97.2*	*95.9*	*90.7*	*96.4*	43.3	*95.3*	*91.8*
W50	76.7	77.2	75.6	79.6	77.1	38	76.6	74.7
W36	78.3	78.6	76.6	80	78.3	38.5	78.2	75.5
W32	44.1	44.7	44.5	42.5	44.6	92.7	44.5	43.1
W31	79.2	79.6	78.1	80.1	79.1	37.5	78.7	75.7
W29	87.4	86.5	86.6	90.1	86.7	40	86.1	84.7
W25	41.7	41.6	42.5	40.7	41.6	*89.7*	42.3	40.9
W24	44	44.6	44.5	42.6	44.5	*92.9*	44.4	43.1
W10	43.4	44	43.9	42.1	44.3	*93.5*	44	42.6
W8	71.6	71.2	70.7	74.4	70.9	36.7	71.3	68.8
S43	78.7	78.2	79.8	80.1	78.1	39.6	78.6	78.4
S42	71.3	71.7	70.2	71.7	71.8	35.8	71.6	69.1
S40	40.2	40.9	40.6	40.3	40.6	77.9	40.6	39.5
S29	*95.5*	*96.3*	*94.1*	*90.4*	*96.7*	43.6	*94.8*	*91*
S26	43	42.8	43.3	44.6	42.6	83.8	43.2	42.1
S23	*95.4*	*95.6*	*93.1*	*90.1*	*96.1*	43.5	*95.3*	*91*
S17	*95.4*	*95.6*	*93.2*	*90.4*	*95.9*	43.2	*95.1*	*91.5*
S16	*95.4*	*95.6*	*93.2*	*90.4*	*95.9*	43.2	*95.1*	*91.5*
S6	*91.1*	*92*	*90*	*93.8*	*91.9*	41.5	*89.9*	86.6
S5	*95.7*	*96.6*	*94.4*	*91.1*	*96.6*	43.2	*94.5*	*91.2*
S4	*95*	*95.5*	*93.1*	*89.5*	*95.7*	43.4	*94.6*	*91*
S3	*94.6*	*95*	*92.5*	*89.5*	*95.3*	43.1	*94.1*	*90.5*

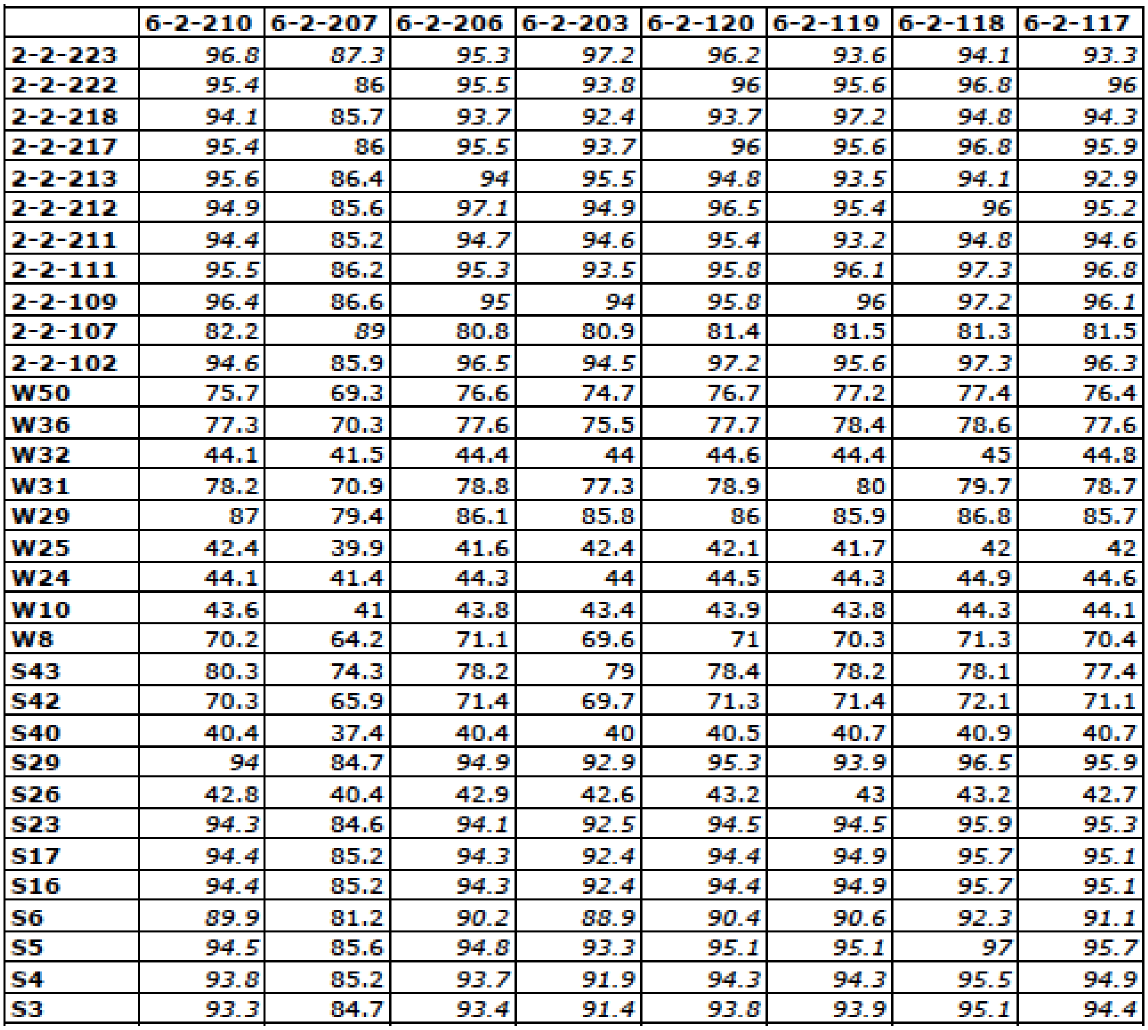
	6-2-406	6-2-405	6-2-401	6-2-310	6-2-309	6-2-306	6-2-304	6-2-303
6-2-510	94	95.7	94.4	92.6	93.5	95	94.4	94.7
6-2-508	94.3	95.9	95	92.8	93.7	94.8	94.5	95.6
6-2-507	96.5	94.1	94.4	92.8	93.7	95.8	97.3	96.3
6-2-504	91.4	89	89	87.9	88.6	90.5	92.3	91
6-2-502	93.5	95.6	93.9	92.4	92.8	94.3	93.9	95.3
6-2-501	43.3	43.3	43	42.3	42.6	43.3	43.9	43.5
6-2-415	94	95.5	93.7	92.2	92.5	95	94.5	94.1
6-2-409	92.6	91.4	92	90	90.9	92.6	93.3	93
6-2-406	**100**	94.5	94.1	93.2	93.4	96.6	96.7	95.9
6-2-405	94.5	**100**	94.9	92.6	92.9	95.2	94.5	94.3
6-2-401	94.1	94.9	**100**	92.7	93.6	94.5	94	94.4
6-2-310	93.2	92.6	92.7	**100**	**99.1**	93.1	93	92.9
6-2-309	93.4	92.9	93.6	**99.1**	**100**	93.3	93.2	93.3
6-2-306	96.6	95.2	94.5	93.1	93.3	**100**	95.5	95.8
6-2-304	96.7	94.5	94	93	93.2	95.5	**100**	96.2
6-2-303	95.9	94.3	94.4	92.9	93.3	95.8	96.2	**100**
6-2-210	97	95.5	95	94.1	94.1	98.1	96.1	96.2
6-2-207	87.3	86.3	86.5	84.6	85.1	87.8	86.6	86.9
6-2-206	95	95.5	93.1	91.6	91.9	94.1	95.8	94.5
6-2-203	96.7	93.7	93.4	91.9	92.2	96	96	95
6-2-120	95.1	95.2	93.6	91.5	92.3	94.9	96.1	94.5
6-2-119	93.6	96.8	96.4	91.7	92.5	94.3	93.4	93.5
6-2-118	93.8	96.1	94.6	92.7	93.6	94.7	94.4	95.5
6-2-117	93	95.3	93.6	91.9	92.5	93.9	93.6	94.6
6-2-113	93.7	96.9	96.5	91.8	92.6	94.4	93.5	93.6
6-2-112	93	95.3	93.4	92	92.4	93.9	93.8	94.6
6-2-111	93	95.3	93.4	92	92.4	93.9	93.8	94.6
6-2-110	93.4	96.6	95.8	92.3	92.7	94.3	93.4	93.5
6-2-108	92.9	95.6	93.8	91.7	92.1	93.9	92.8	93.2
6-2-106	95.1	95.5	93.5	92.8	93.2	96.2	94.7	94.8
6-2-105	95.1	95.5	93.5	92.8	93.2	96.2	94.7	94.8
6-2-104	94.3	95.9	94	92.4	92.8	95.7	94.8	95.3
6-2-103	94.9	96.2	94.1	92.6	92.9	94.5	95.7	94.5
2-2-508	94.4	95.7	94	92.1	92.5	93.9	95.2	94.2
2-2-503	94.2	95.1	94.7	93.1	93.2	94.8	94.6	94.2
2-2-501	93.2	95.5	93.8	91.7	92.2	94	93.8	93.8
2-2-418	93.5	96	95	92.5	93.4	94.1	93.9	93.8
2-2-402	93.7	95.7	94.4	92.7	93.6	94.2	93.8	94.1
2-2-330	84	83.3	83.6	82	82.5	83.8	83.4	83.6
2-2-329	93	92.1	93.4	94.8	95.6	93	92.8	93.2
2-2-324	77.6	78.5	78.2	76.6	77.4	78.4	77.8	78.4
2-2-320	95.6	92.9	92.9	91.4	92.3	94.6	95.7	94.7
2-2-318	92.5	95.3	93.9	91.8	92.3	93.5	92.9	93
2-2-317	94.5	94.6	93.7	91.9	92.4	94.8	94.3	94.1
2-2-309	94.9	96.5	94.7	92.9	93.3	96.4	95.1	95.5
2-2-306	96.3	94.1	93.1	93.1	93.3	96	96.2	95.1
2-2-305	91.7	91.4	91.5	92.7	93.1	91.7	92.2	92
2-2-304	92.3	92	91.9	93.5	93.6	92.1	92.6	92.4



	6-2-406	6-2-405	6-2-401	6-2-310	6-2-309	6-2-306	6-2-304	6-2-303
2-2-223	*97.2*	*94.1*	*93.8*	*92.7*	*93.5*	*96.5*	*96.8*	*95.3*
2-2-222	*94.3*	*95.9*	*94.2*	*92.9*	*93.6*	*95.2*	*94.6*	*94.9*
2-2-218	*93*	*96.2*	*95.3*	*91.4*	*91.8*	*93.8*	*92.8*	*92.9*
2-2-217	*94.3*	*95.9*	*94.2*	*92.9*	*93.6*	*95.2*	*94.6*	*94.9*
2-2-213	*95.3*	*93.5*	*92.9*	*91.7*	*92.6*	*94.9*	*95.5*	*94.1*
2-2-212	*95.2*	*96.5*	*93.7*	*92.1*	*92.3*	*94.4*	*95.8*	*94.2*
2-2-211	*94.7*	*93.8*	*94.8*	*92.6*	*93.3*	*93.7*	*95.5*	*94.7*
2-2-111	*94.1*	*96.6*	*95*	*93.2*	*93.9*	*95.1*	*94.4*	*94.4*
2-2-109	*95.1*	*96.4*	*95.1*	*93.9*	*94.8*	*95.9*	*95.4*	*95.4*
2-2-107	81.9	81.8	81.9	80.9	81.6	81.8	81.4	81.5
2-2-102	*94.6*	*95.9*	*94.5*	*92.3*	*93.1*	*94.1*	*95.4*	*94.4*
W50	75.3	77.3	75.7	74	74.5	75.3	75.6	75.2
W36	76.2	78.7	77.2	75.6	76	77	76.8	76.4
W32	43.8	44.5	44.4	43.4	43.6	43.8	44.7	44.4
W31	77.6	79.6	78.4	76.3	76.9	78	77.7	77.6
W29	86	86.2	86.5	84.2	84.8	86.6	86	86.2
W25	41.8	41.8	41.6	40.9	41.1	42.1	42.4	42
W24	43.7	44.4	44.5	43.3	43.6	44	44.7	44.4
W10	43.2	43.9	43.8	42.8	43	43.3	44.1	43.8
W8	70	70.9	69.5	67.6	68.1	70	70.5	70.5
S43	79.9	78.4	77.6	76.9	77.4	79.5	79.6	79
S42	69.6	71.6	70.3	68.4	68.8	70.4	70.4	69.9
S40	40.2	40.3	40.4	39.2	39.6	40.2	40.7	40.7
S29	93	*94.9*	*93.5*	*91.6*	*92.1*	*93.8*	*94.1*	*94.2*
S26	42.6	42.9	42.5	41.5	41.7	42.5	43.5	43.2
S23	*92.9*	*95.4*	*93.5*	*91.6*	*92.1*	*94.1*	*93.3*	*93.6*
S17	*92.9*	*95.8*	*93.7*	*91.6*	*92.1*	*94.1*	*93.4*	*93.6*
S16	*92.9*	*95.8*	*93.7*	*91.6*	*92.1*	*94.1*	*93.4*	*93.6*
S6	*89.1*	*90.7*	*89.8*	*87.5*	*88.2*	*89.8*	*89.5*	*90*
S5	*93.6*	*95.3*	*94.4*	*92.2*	*92.8*	*94.4*	*94.1*	*94.7*
S4	*92.6*	*95.3*	*93.6*	*91.7*	*92.1*	*93.6*	*93*	*93.3*
S3	*92.1*	*94.9*	*93*	*91.3*	*91.8*	*93.1*	*92.5*	*92.9*



	6-2-210	**6-2-207**	**6-2-206**	**6-2-203**	**6-2-120**	**6-2-119**	**6-2-118**	**6-2-117**
6-2-510	*95.4*	86.8	*95.1*	*93.4*	*95.9*	*95.2*	*96.8*	*95.7*
6-2-508	*95*	86	*95.4*	*93.5*	*95.7*	*95.7*	*98.1*	*97.1*
6-2-507	*96.3*	*87.3*	*95.3*	*96.6*	*96.1*	*93.7*	*94.8*	*93.8*
6-2-504	*91.1*	82.5	*90.4*	*91.7*	*90.7*	*89.1*	*89.6*	*88.4*
6-2-502	*94.5*	85.4	*95.2*	*93.2*	*95.2*	*94.8*	*97.5*	*97.1*
6-2-501	43.6	41	43	43.5	43.5	43.2	43.3	43.5
6-2-415	*95.6*	85.5	*94.8*	*93.2*	*95.4*	*94.4*	*95.8*	*95*
6-2-409	*93.3*	86.3	*91.2*	*91.8*	*91.9*	*91.3*	*91.8*	*91*
6-2-406	*97*	*87.3*	*95*	*96.7*	*95.1*	*93.6*	*93.8*	*93*
6-2-405	*95.5*	86.3	*95.5*	*93.7*	*95.2*	*96.8*	*96.1*	*95.3*
6-2-401	*95*	86.5	*93.1*	*93.4*	*93.6*	*96.4*	*94.6*	*93.6*
6-2-310	*94.1*	84.6	*91.6*	*91.9*	*91.5*	*91.7*	*92.7*	*91.9*
6-2-309	*94.1*	85.1	*91.9*	*92.2*	*92.3*	*92.5*	*93.6*	*92.5*
6-2-306	*98.1*	*87.8*	*94.1*	*96*	*94.9*	*94.3*	*94.7*	*93.9*
6-2-304	*96.1*	86.6	*95.8*	*96*	*96.1*	*93.4*	*94.4*	*93.6*
6-2-303	*96.2*	86.9	*94.5*	*95*	*94.5*	*93.5*	*95.5*	*94.6*
6-2-210	**100**	*88.3*	*94.2*	*96.2*	*94.4*	*94.6*	*95.1*	*94.2*
6-2-207	*88.3*	**100**	85.5	*87.3*	85.8	86.4	85.8	85.2
6-2-206	*94.2*	85.5	**100**	*94.6*	*96.7*	*94.5*	*95.5*	*94.6*
6-2-203	*96.2*	*87.3*	*94.6*	**100**	*95.2*	*93.1*	*93.5*	*92.7*
6-2-120	*94.4*	85.8	*96.7*	*95.2*	**100**	*94.6*	*96.1*	*95.1*
6-2-119	*94.6*	86.4	*94.5*	*93.1*	*94.6*	**100**	*95.9*	*94.7*
6-2-118	*95.1*	85.8	*95.5*	*93.5*	*96.1*	*95.9*	**100**	*97.6*
6-2-117	*94.2*	85.2	*94.6*	*92.7*	*95.1*	*94.7*	*97.6*	**100**
6-2-113	*94.7*	86.5	*94.6*	*93.2*	*94.7*	**99.5**	*96*	*94.8*
6-2-112	*94.2*	84.8	*94.7*	*92.7*	*94.9*	*94.5*	*97.3*	*96.8*
6-2-111	*94.2*	84.8	*94.7*	*92.7*	*94.9*	*94.5*	*97.3*	*96.8*
6-2-110	*94.6*	86.3	*94.3*	*93.1*	*94.6*	*97.6*	*95.8*	*94.9*
6-2-108	*94.2*	85.5	*93.8*	*92.3*	*94.1*	*94.7*	*95.6*	*94.7*
6-2-106	*96.4*	86.5	*94.3*	*93.9*	*94.7*	*94.6*	*95.1*	*94.2*
6-2-105	*96.4*	86.5	*94.3*	*93.9*	*94.7*	*94.6*	*95.1*	*94.2*
6-2-104	*96.1*	85.9	*95.2*	*93.7*	*95.9*	*95.2*	*96.7*	*95.8*
6-2-103	*95.2*	85.9	*96.9*	*94.2*	*96.7*	*95.7*	*97.1*	*96*
2-2-508	*94.4*	85.6	*96.3*	*94.3*	*96.6*	*95.1*	*96.8*	*95.9*
2-2-503	*95.6*	86.2	*94.7*	*93.5*	*94.7*	*94.5*	*95.2*	*94.4*
2-2-501	*94.3*	85.6	*95*	*93*	*94.9*	*94.7*	*96.1*	*95.3*
2-2-418	*94.5*	85.5	*95.2*	*93*	*95.4*	*95.8*	*96.7*	*95.7*
2-2-402	*94.4*	85.3	*94.9*	*92.8*	*95.2*	*95.5*	*96.8*	*95.7*
2-2-330	84.1	85.6	82.4	83.1	82.5	82.8	82.4	82.5
2-2-329	*93.3*	85.3	*92.2*	*92.2*	*92.2*	*92.2*	*92.9*	*91.9*
2-2-324	78.7	75.5	77.7	77.2	79	78.3	79.6	78.9
2-2-320	*95.3*	86.6	*93.9*	*95.5*	*94.5*	*93*	*93.6*	*92.5*
2-2-318	*93.9*	85.1	*93.7*	*92*	*94*	*94.3*	*95.5*	*95*
2-2-317	*94.3*	84.9	*94.1*	*93.5*	*94.4*	*93.9*	*94.7*	*94.3*
2-2-309	*96.6*	86.5	*95.7*	*94.3*	*96.3*	*95.7*	*96.9*	*96*
2-2-306	*96.6*	*87.2*	*94.7*	*95.9*	*94.8*	*93.1*	*93.8*	*93*
2-2-305	*92.2*	83.7	*91.4*	*91.3*	*91.1*	*90.9*	*91.6*	*91.3*
2-2-304	*93*	84.2	*91.9*	*91.5*	*91.4*	*91.3*	*92*	*91.3*



	6-2-210	6-2-207	6-2-206	6-2-203	6-2-120	6-2-119	6-2-118	6-2-117
2-2-223	*96.8*	*87.3*	*95.3*	*97.2*	*96.2*	*93.6*	*94.1*	*93.3*
2-2-222	*95.4*	86	*95.5*	*93.8*	*96*	*95.6*	*96.8*	*96*
2-2-218	*94.1*	85.7	*93.7*	*92.4*	*93.7*	*97.2*	*94.8*	*94.3*
2-2-217	*95.4*	86	*95.5*	*93.7*	*96*	*95.6*	*96.8*	*95.9*
2-2-213	*95.6*	86.4	*94*	*95.5*	*94.8*	*93.5*	*94.1*	*92.9*
2-2-212	*94.9*	85.6	*97.1*	*94.9*	*96.5*	*95.4*	*96*	*95.2*
2-2-211	*94.4*	85.2	*94.7*	*94.6*	*95.4*	*93.2*	*94.8*	*94.6*
2-2-111	*95.5*	86.2	*95.3*	*93.5*	*95.8*	*96.1*	*97.3*	*96.8*
2-2-109	*96.4*	86.6	*95*	*94*	*95.8*	*96*	*97.2*	*96.1*
2-2-107	82.2	*89*	80.8	80.9	81.4	81.5	81.3	81.5
2-2-102	*94.6*	85.9	*96.5*	*94.5*	*97.2*	*95.6*	*97.3*	*96.3*
W50	75.7	69.3	76.6	74.7	76.7	77.2	77.4	76.4
W36	77.3	70.3	77.6	75.5	77.7	78.4	78.6	77.6
W32	44.1	41.5	44.4	44	44.6	44.4	45	44.8
W31	78.2	70.9	78.8	77.3	78.9	80	79.7	78.7
W29	87	79.4	86.1	85.8	86	85.9	86.8	85.7
W25	42.4	39.9	41.6	42.4	42.1	41.7	42	42
W24	44.1	41.4	44.3	44	44.5	44.3	44.9	44.6
W10	43.6	41	43.8	43.4	43.9	43.8	44.3	44.1
W8	70.2	64.2	71.1	69.6	71	70.3	71.3	70.4
S43	80.3	74.3	78.2	79	78.4	78.2	78.1	77.4
S42	70.3	65.9	71.4	69.7	71.3	71.4	72.1	71.1
S40	40.4	37.4	40.4	40	40.5	40.7	40.9	40.7
S29	*94*	84.7	*94.9*	*92.9*	*95.3*	*93.9*	*96.5*	*95.9*
S26	42.8	40.4	42.9	42.6	43.2	43	43.2	42.7
S23	*94.3*	84.6	*94.1*	*92.5*	*94.5*	*94.5*	*95.9*	*95.3*
S17	*94.4*	85.2	*94.3*	*92.4*	*94.4*	*94.9*	*95.7*	*95.1*
S16	*94.4*	85.2	*94.3*	*92.4*	*94.4*	*94.9*	*95.7*	*95.1*
S6	*89.9*	81.2	*90.2*	*88.9*	*90.4*	*90.6*	*92.3*	*91.1*
S5	*94.5*	85.6	*94.8*	*93.3*	*95.1*	*95.1*	*97*	*95.7*
S4	*93.8*	85.2	*93.7*	*91.9*	*94.3*	*94.3*	*95.5*	*94.9*
S3	*93.3*	84.7	*93.4*	*91.4*	*93.8*	*93.9*	*95.1*	*94.4*

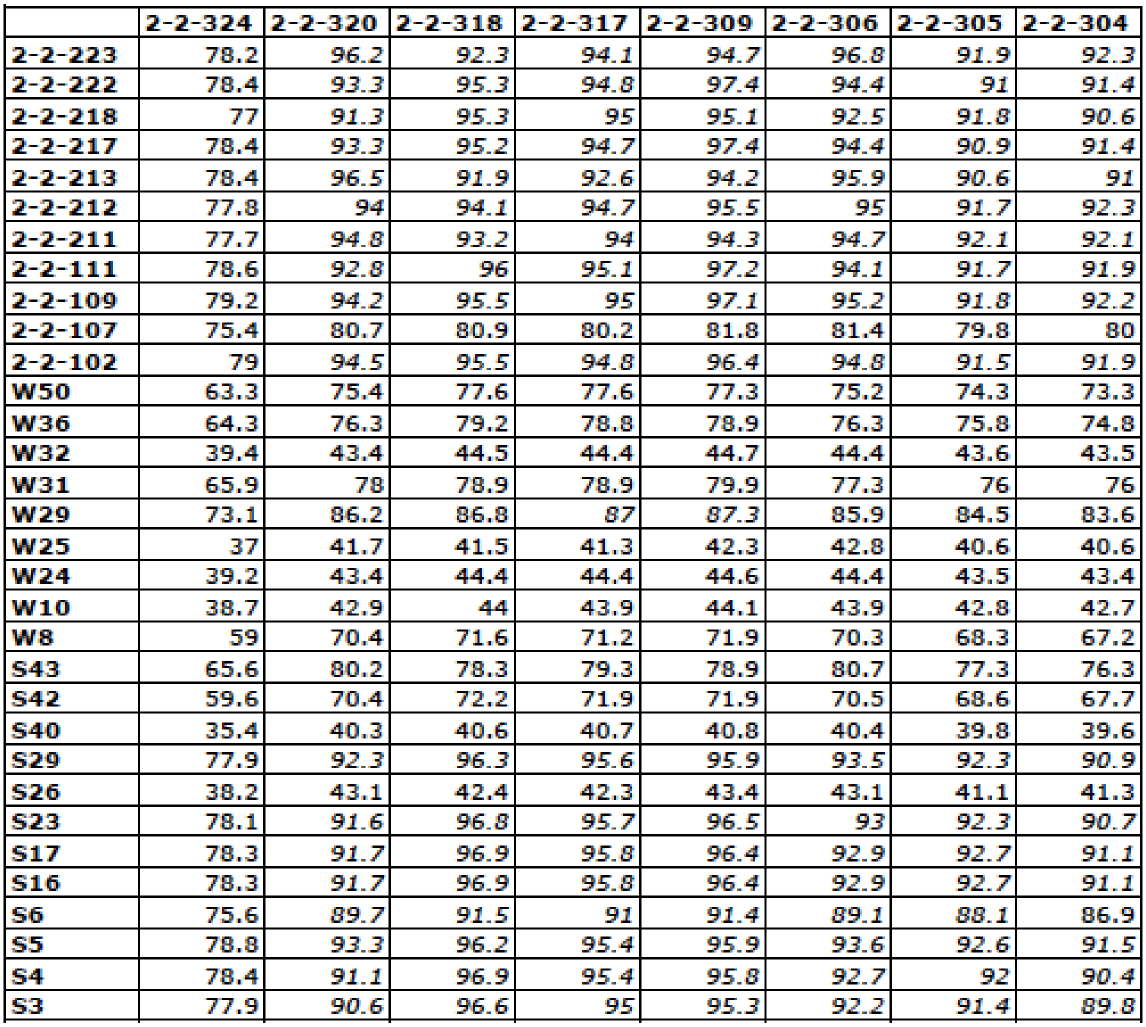
	6-2-113	6-2-112	6-2-111	6-2-110	6-2-108	6-2-106	6-2-105	6-2-104
6-2-510	95.3	95.6	95.6	95.4	95.4	95.6	95.6	96.7
6-2-508	95.8	96.9	96.9	95.5	95.4	95.3	95.3	96.3
6-2-507	93.8	93.6	93.6	93.3	92.8	94.9	94.9	94.7
6-2-504	89	90.4	90.4	88.5	88.9	89.8	89.8	89.6
6-2-502	94.9	97.7	97.7	95.4	95.5	94.9	94.9	96.1
6-2-501	43.3	43.6	43.6	43.8	43.1	43.8	43.8	43.7
6-2-415	94.5	94.8	94.8	95	94.7	95.8	95.8	96.8
6-2-409	91.4	90.6	90.6	90.8	90.9	92.7	92.7	92.1
6-2-406	93.7	93	93	93.4	92.9	95.1	95.1	94.3
6-2-405	96.9	95.3	95.3	96.6	95.6	95.5	95.5	95.9
6-2-401	96.5	93.4	93.4	95.8	93.8	93.5	93.5	94
6-2-310	91.8	92	92	92.3	91.7	92.8	92.8	92.4
6-2-309	92.6	92.4	92.4	92.7	92.1	93.2	93.2	92.8
6-2-306	94.4	93.9	93.9	94.3	93.9	96.2	96.2	95.7
6-2-304	93.5	93.8	93.8	93.4	92.8	94.7	94.7	94.8
6-2-303	93.6	94.6	94.6	93.5	93.2	94.8	94.8	95.3
6-2-210	94.7	94.2	94.2	94.6	94.2	96.4	96.4	96.1
6-2-207	86.5	84.8	84.8	86.3	85.5	86.5	86.5	85.9
6-2-206	94.6	94.7	94.7	94.3	93.8	94.3	94.3	95.2
6-2-203	93.2	92.7	92.7	93.1	92.3	93.9	93.9	93.7
6-2-120	94.7	94.9	94.9	94.6	94.1	94.7	94.7	95.9
6-2-119	**99.5**	94.5	94.5	97.6	94.7	94.6	94.6	95.2
6-2-118	96	97.3	97.3	95.8	95.6	95.1	95.1	96.7
6-2-117	94.8	96.8	96.8	94.9	94.7	94.2	94.2	95.8
6-2-113	**100**	94.6	94.6	97.7	94.7	94.7	94.7	95.3
6-2-112	94.6	**100**	**99.8**	95	95.5	94.3	94.3	95.8
6-2-111	94.6	**99.8**	**100**	95	95.5	94.3	94.3	95.8
6-2-110	97.7	95	95	**100**	95.1	94.8	94.8	95.9
6-2-108	94.7	95.5	95.5	95.1	**100**	94.2	94.2	95.3
6-2-106	94.7	94.3	94.3	94.8	94.2	**100**	**100**	96.3
6-2-105	94.7	94.3	94.3	94.8	94.2	**100**	**100**	96.3
6-2-104	95.3	95.8	95.8	95.9	95.3	96.3	96.3	**100**
6-2-103	95.8	96	96	95.8	95.1	95.2	95.2	96.4
2-2-508	95.2	95.9	95.9	95.3	95.3	94.6	94.6	95.8
2-2-503	94.6	94.3	94.3	94.7	94	95.1	95.1	95.4
2-2-501	94.8	95.4	95.4	95.1	94.9	94.7	94.7	95.5
2-2-418	95.9	95.5	95.5	95.5	95.3	94.6	94.6	95.7
2-2-402	95.5	95.6	95.6	95.4	95.1	95	95	95.7
2-2-330	82.9	83.6	83.6	82.7	83.2	83.6	83.6	83.1
2-2-329	92.3	91.7	91.7	91.6	91.3	92.2	92.2	92.6
2-2-324	78.3	77.7	77.7	77.7	78.2	78.2	78.2	78.9
2-2-320	92.9	92.2	92.2	91.8	91.7	93.9	93.9	93.3
2-2-318	94.3	96.6	96.6	94.8	96.4	94.1	94.1	95.2
2-2-317	94	96.2	96.2	93.9	94.5	93.8	93.8	94.2
2-2-309	95.8	96	96	96	96	97.1	97.1	98
2-2-306	93.2	93.3	93.3	93.6	92.6	95.3	95.3	94.4
2-2-305	91	93	93	90.7	91.1	90.7	90.7	91.1
2-2-304	91.4	91.2	91.2	91.1	90.7	91.1	91.1	91.5

	6-2-113	**6-2-112**	**6-2-111**	**6-2-110**	**6-2-108**	**6-2-106**	**6-2-105**	**6-2-104**
2-2-223	*93.7*	*93.1*	*93.1*	*93.4*	*92.6*	*94.4*	*94.4*	*94.4*
2-2-222	*95.7*	*95.8*	*95.8*	*95.6*	*95.4*	*95.7*	*95.7*	*96.9*
2-2-218	*97.3*	*95.6*	*95.6*	*97.2*	*94.7*	*94.3*	*94.3*	*94.6*
2-2-217	*95.7*	*95.7*	*95.7*	*95.6*	*95.4*	*95.7*	*95.7*	*96.9*
2-2-213	*93.4*	*92.5*	*92.5*	*92.6*	*92.1*	*94.1*	*94.1*	*93.9*
2-2-212	*95.5*	*95.2*	*95.2*	*95*	*94.2*	*94.5*	*94.5*	*95.2*
2-2-211	*93.3*	*94*	*94*	*93.5*	*92.8*	*93.1*	*93.1*	*94.1*
2-2-111	*96.2*	*96.5*	*96.5*	*96.5*	*95.7*	*95.8*	*95.8*	*96.7*
2-2-109	*96.1*	*96*	*96*	*96*	*95.4*	*97.2*	*97.2*	*96.8*
2-2-107	81.6	80.4	80.4	81.4	80.7	82	82	81.4
2-2-102	*95.7*	*96.1*	*96.1*	*95.5*	*95.4*	*94.8*	*94.8*	*96*
W50	77.2	78.1	78.1	77	78.2	76.3	76.3	77.1
W36	78.5	79.3	79.3	78.3	79.5	78	78	78.5
W32	44.5	44.9	44.9	44.8	44.8	44.5	44.5	44.4
W31	79.9	79	79	79.7	79.9	78.5	78.5	79.5
W29	85.8	*87.2*	*87.2*	85.8	86.6	86.4	86.4	86.5
W25	41.8	41.9	41.9	42.2	41.6	42.1	42.1	42.2
W24	44.4	44.8	44.8	44.9	44.7	44.5	44.5	44.5
W10	43.8	44.3	44.3	44.3	44.1	44.1	44.1	44
W8	70.2	72.1	72.1	70.1	71.1	71	71	71.2
S43	78.2	79	79	77.8	78.2	79.2	79.2	78.7
S42	71.5	72.9	72.9	71.4	72.6	70.7	70.7	71.6
S40	40.7	41.4	41.4	40.9	40.8	40.7	40.7	41
S29	*94*	*97.5*	*97.5*	*94.4*	*95.5*	*94.3*	*94.3*	*95.5*
S26	42.9	42.8	42.8	43	43	43.4	43.4	43.2
S23	*94.6*	*97.3*	*97.3*	*95*	*96.2*	*94.8*	*94.8*	*95.9*
S17	*95*	*97.1*	*97.1*	*95*	*96.7*	*94.9*	*94.9*	*95.7*
S16	*95*	*97.1*	*97.1*	*95*	*96.7*	*94.9*	*94.9*	*95.7*
S6	*90.5*	*92.8*	*92.8*	*90.2*	*91.1*	*90.1*	*90.1*	*90.9*
S5	*95.1*	*97.2*	*97.2*	*94.7*	*95.3*	*94.7*	*94.7*	*95.4*
S4	*94.4*	*96.7*	*96.7*	*94.9*	*96.9*	*94.2*	*94.2*	*95.3*
S3	*94*	*96.5*	*96.5*	*94.5*	*96.5*	*93.8*	*93.8*	*94.8*

	6-2-103	2-2-508	2-2-503	2-2-501	2-2-418	2-2-402	2-2-330	2-2-329
6-2-510	95.9	95.8	95.9	96.1	96.7	96	82.8	92.9
6-2-508	96.5	96.6	95.1	95.9	96.5	97	82.6	92.9
6-2-507	95.1	95.4	94.6	93.8	94.3	94	84	93.4
6-2-504	90.1	90.4	89.6	89.1	89.1	88.7	80.4	88.1
6-2-502	96.3	96.2	94.6	95.8	95.7	96	82.8	92.2
6-2-501	43.5	43.4	42.9	43.3	43.1	42.7	40.1	41.8
6-2-415	95.8	95.2	95.4	94.8	95.1	95.3	82.5	91.5
6-2-409	91.8	91.3	91.8	91.2	91.6	91.3	83.1	91.1
6-2-406	94.9	94.4	94.2	93.2	93.5	93.7	84	93
6-2-405	96.2	95.7	95.1	95.5	96	95.7	83.3	92.1
6-2-401	94.1	94	94.7	93.8	95	94.4	83.6	93.4
6-2-310	92.6	92.1	93.1	91.7	92.5	92.7	82	94.8
6-2-309	92.9	92.5	93.2	92.2	93.4	93.6	82.5	95.6
6-2-306	94.5	93.9	94.8	94	94.1	94.2	83.8	93
6-2-304	95.7	95.2	94.6	93.8	93.9	93.8	83.4	92.8
6-2-303	94.5	94.2	94.2	93.8	93.8	94.1	83.6	93.2
6-2-210	95.2	94.4	95.6	94.3	94.5	94.4	84.1	93.3
6-2-207	85.9	85.6	86.2	85.6	85.5	85.3	85.6	85.3
6-2-206	96.9	96.3	94.7	95	95.2	94.9	82.4	92.2
6-2-203	94.2	94.3	93.5	93	93	92.8	83.1	92.2
6-2-120	96.7	96.6	94.7	94.9	95.4	95.2	82.5	92.2
6-2-119	95.7	95.1	94.5	94.7	95.8	95.5	82.8	92.2
6-2-118	97.1	96.8	95.2	96.1	96.7	96.8	82.4	92.9
6-2-117	96	95.9	94.4	95.3	95.7	95.7	82.5	91.9
6-2-113	95.8	95.2	94.6	94.8	95.9	95.5	82.9	92.3
6-2-112	96	95.9	94.3	95.4	95.5	95.6	83.6	91.7
6-2-111	96	95.9	94.3	95.4	95.5	95.6	83.6	91.7
6-2-110	95.8	95.3	94.7	95.1	95.5	95.4	82.7	91.6
6-2-108	95.1	95.3	94	94.9	95.3	95.1	83.2	91.3
6-2-106	95.2	94.6	95.1	94.7	94.6	95	83.6	92.2
6-2-105	95.2	94.6	95.1	94.7	94.6	95	83.6	92.2
6-2-104	96.4	95.8	95.4	95.5	95.7	95.7	83.1	92.6
6-2-103	100	97.3	95.6	96.2	96.4	96.3	82.7	92.8
2-2-508	97.3	100	94.8	95.8	96.3	96	82.4	92.4
2-2-503	95.6	94.8	100	94.9	95.8	95.1	82.7	92.7
2-2-501	96.2	95.8	94.9	100	96	95.2	82.4	91.8
2-2-418	96.4	96.3	95.8	96	100	96.5	81.9	92.6
2-2-402	96.3	96	95.1	95.2	96.5	100	82	92.3
2-2-330	82.7	82.4	82.7	82.4	81.9	82	100	82.7
2-2-329	92.8	92.4	92.7	91.8	92.6	92.3	82.7	100
2-2-324	78.4	78.5	78.6	78	78.3	78.2	75.7	77.5
2-2-320	93.7	94	92.8	92.4	92.9	92.4	82.4	92
2-2-318	95.1	95.3	94.4	95.3	95.7	94.8	83.6	91.1
2-2-317	94.8	94.6	93.5	93.9	94.3	94.3	83.8	91.6
2-2-309	96.8	96.2	95.9	96	96.2	96.5	83.6	92.8
2-2-306	94.6	94.6	94.1	93.4	93.7	93.4	83.3	92.6
2-2-305	92	91.4	91.4	90.8	90.8	90.9	82.9	93.8
2-2-304	92.6	91.7	92	91.2	91.3	91.4	81.3	94.2

	6-2-103	**2-2-508**	**2-2-503**	**2-2-501**	**2-2-418**	**2-2-402**	**2-2-330**	**2-2-329**
2-2-223	*95.1*	*95.1*	*94.2*	*93.6*	*93.9*	*93.5*	83.3	*93*
2-2-222	*96.5*	*96.1*	*95.8*	*95.5*	*96.8*	*96.7*	82.9	*92.6*
2-2-218	*94.9*	*94.4*	*94.1*	*94*	*94.5*	*94.4*	83.6	*90.9*
2-2-217	*96.5*	*96.3*	*95.8*	*95.5*	*96.8*	*96.7*	82.8	*92.6*
2-2-213	*94*	*94.1*	*93.1*	*93*	*93.5*	*93*	82.6	*92.4*
2-2-212	*97.3*	*96.9*	*94.7*	*95.4*	*95.5*	*95.4*	82.3	*92.4*
2-2-211	*95.1*	*95.4*	*93.9*	*93.8*	*94.2*	*93.7*	83	*93.3*
2-2-111	*97.1*	*96.2*	*95.8*	*96.4*	*96.8*	*96.9*	83.4	*92.6*
2-2-109	*96.6*	*95.9*	*95.8*	*96.1*	*96.4*	*96.2*	83.6	*93.9*
2-2-107	81.4	81	81.6	81.1	80.8	80.5	*94.4*	81.2
2-2-102	*97.4*	**99.4**	*95*	*96*	*96.9*	*96.6*	82.6	*92.9*
W50	77.7	77.7	76	76.8	77.5	77.6	67.7	73.8
W36	78.6	78.6	77.7	78.4	79	78.9	68.6	75.4
W32	44.9	44.7	44.1	44.8	44.6	44.1	40.4	42.7
W31	80	79.6	78.5	79.2	79.9	79.7	68.8	76.7
W29	86.5	86	*88.1*	86.4	86.6	86.2	76.7	84.4
W25	41.9	41.8	41.4	41.7	41.6	41.2	38.2	40.3
W24	44.8	44.6	44.1	44.6	44.5	44	40.2	42.9
W10	44.3	44.3	43.5	44.1	43.9	43.5	39.8	42.3
W8	70.6	71.1	70.6	71.2	70.9	70.9	62.6	67.6
S43	78.3	78.1	78.4	77.6	78.2	78.3	72	77.2
S42	72.1	72.3	70.7	71.4	71.5	71.8	63.7	69.3
S40	41	40.8	40.2	40.8	40.8	40.4	36.4	39.2
S29	*95.8*	*95.8*	*94.5*	*95.5*	*95.5*	*95.1*	83.3	*91.7*
S26	43.1	43	42.6	42.9	42.8	42.5	38.7	40.8
S23	*95.5*	*95.2*	*94.3*	*95.1*	*95.2*	*95.3*	84	*91.9*
S17	*95.6*	*95.1*	*94.3*	*95.3*	*95.3*	*95.5*	84.2	*91.5*
S16	*95.6*	*95.1*	*94.3*	*95.3*	*95.3*	*95.5*	84.2	*91.5*
S6	*91.3*	*91.2*	*90.5*	*91.1*	*91.1*	*91*	79	*87.5*
S5	*95.9*	*95.6*	*95*	*95.4*	*95.7*	*95.6*	83.2	*92.1*
S4	*94.9*	*95.1*	*93.9*	*94.5*	*95*	*95*	84.5	*91.3*
S3	*94.4*	*94.6*	*93.4*	*94.1*	*94.5*	*94.4*	83.9	*90.7*

	2-2-324	2-2-320	2-2-318	2-2-317	2-2-309	2-2-306	2-2-305	2-2-304
6-2-510	78.7	93.2	96.1	94.3	97.3	94.2	91	91.4
6-2-508	78.9	93.4	95.3	94.7	96.8	93.7	91.6	92
6-2-507	79.1	97.1	92.7	94	95.2	96.7	92	92.4
6-2-504	75.2	93.2	89.1	90.8	90	91.5	88.7	87.3
6-2-502	78.2	92.5	96	95.2	96.5	93.7	92	91.7
6-2-501	38	42.9	43.3	43.1	43.8	44	42.2	42
6-2-415	78.1	92.5	94.5	93.5	97.5	94.1	90.3	90.9
6-2-409	78.7	91.8	90.4	90.3	92.7	92.4	89	89.3
6-2-406	77.6	95.6	92.5	94.5	94.9	96.3	91.7	92.3
6-2-405	78.5	92.9	95.3	94.6	96.5	94.1	91.4	92
6-2-401	78.2	92.9	93.9	93.7	94.7	93.1	91.5	91.9
6-2-310	76.6	91.4	91.8	91.9	92.9	93.1	92.7	93.5
6-2-309	77.4	92.3	92.3	92.4	93.3	93.3	93.1	93.6
6-2-306	78.4	94.6	93.5	94.8	96.4	96	91.7	92.1
6-2-304	77.8	95.7	92.9	94.3	95.1	96.2	92.2	92.6
6-2-303	78.4	94.7	93	94.1	95.5	95.1	92	92.4
6-2-210	78.7	95.3	93.9	94.3	96.6	96.6	92.2	93
6-2-207	75.5	86.6	85.1	84.9	86.5	87.2	83.7	84.2
6-2-206	77.7	93.9	93.7	94.1	95.7	94.7	91.4	91.9
6-2-203	77.2	95.5	92	93.5	94.3	95.9	91.3	91.5
6-2-120	79	94.5	94	94.4	96.3	94.8	91.1	91.4
6-2-119	78.3	93	94.3	93.9	95.7	93.1	90.9	91.3
6-2-118	79.6	93.6	95.5	94.7	96.9	93.8	91.6	92
6-2-117	78.9	92.5	95	94.3	96	93	91.3	91.3
6-2-113	78.3	92.9	94.3	94	95.8	93.2	91	91.4
6-2-112	77.7	92.2	96.6	96.2	96	93.3	93	91.2
6-2-111	77.7	92.2	96.6	96.2	96	93.3	93	91.2
6-2-110	77.7	91.8	94.8	93.9	96	93.6	90.7	91.1
6-2-108	78.2	91.7	96.4	94.5	96	92.6	91.1	90.7
6-2-106	78.2	93.9	94.1	93.8	97.1	95.3	90.7	91.1
6-2-105	78.2	93.9	94.1	93.8	97.1	95.3	90.7	91.1
6-2-104	78.9	93.3	95.2	94.2	98	94.4	91.1	91.5
6-2-103	78.4	93.7	95.1	94.8	96.8	94.6	92	92.6
2-2-508	78.5	94	95.3	94.6	96.2	94.6	91.4	91.7
2-2-503	78.6	92.8	94.4	93.5	95.9	94.1	91.4	92
2-2-501	78	92.4	95.3	93.9	96	93.4	90.8	91.2
2-2-418	78.3	92.9	95.7	94.3	96.2	93.7	90.8	91.3
2-2-402	78.2	92.4	94.8	94.3	96.5	93.4	90.9	91.4
2-2-330	75.7	82.4	83.6	83.8	83.6	83.3	82.9	81.3
2-2-329	77.5	92	91.1	91.6	92.8	92.6	93.8	94.2
2-2-324	100	78.9	77.9	77.2	79.2	77.3	76.2	76.7
2-2-320	78.9	100	91.3	92.7	93.6	95.5	90.6	91
2-2-318	77.9	91.3	100	95.3	95.8	92.7	91.8	90.4
2-2-317	77.2	92.7	95.3	100	94.6	93.6	92.7	90.9
2-2-309	79.2	93.6	95.8	94.6	100	95.1	91.7	92.1
2-2-306	77.3	95.5	92.7	93.6	95.1	100	90.9	91.4
2-2-305	76.2	90.6	91.8	92.7	91.7	90.9	100	97.6
2-2-304	76.7	91	90.4	90.9	92.1	91.4	97.6	100

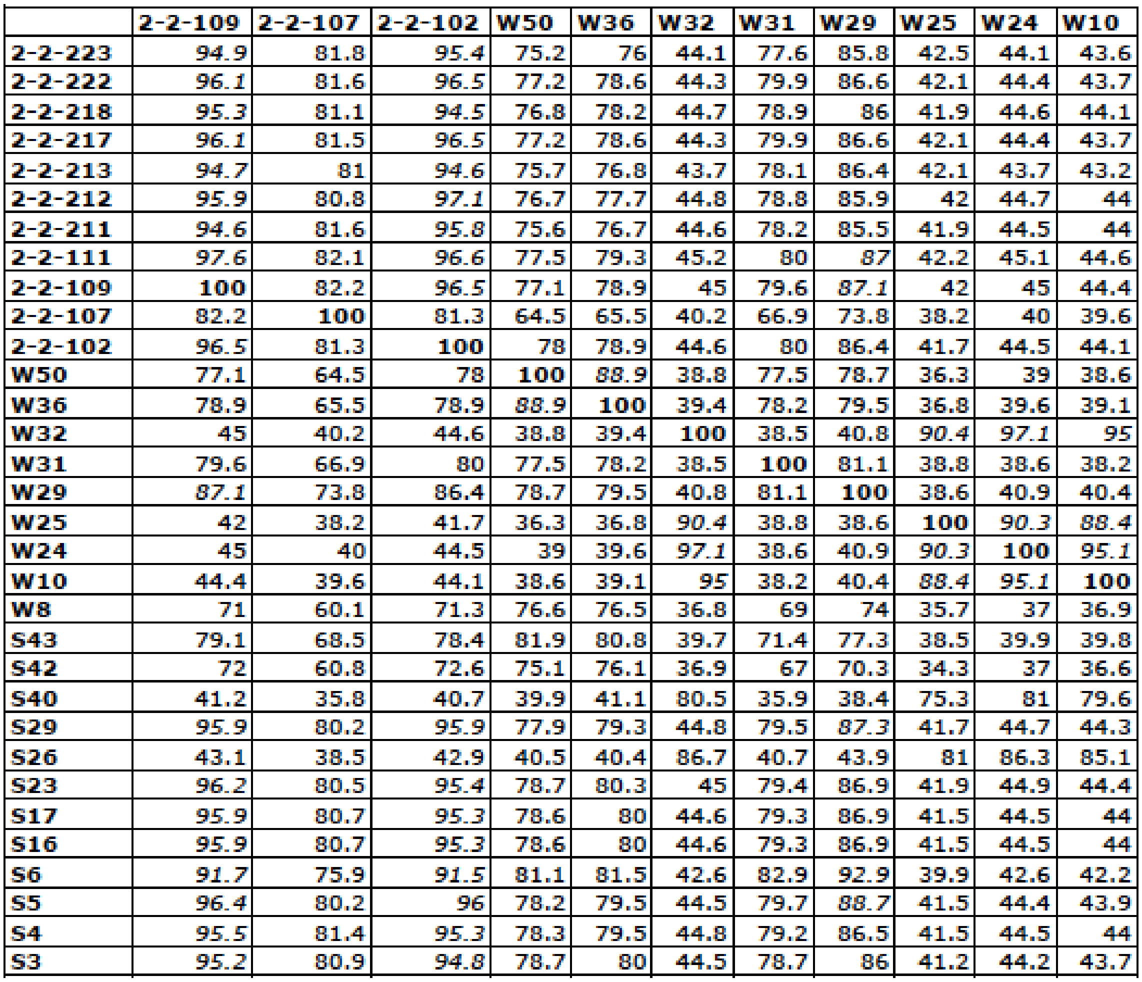


	2-2-324	**2-2-320**	**2-2-318**	**2-2-317**	**2-2-309**	**2-2-306**	**2-2-305**	**2-2-304**
2-2-223	78.2	*96.2*	*92.3*	*94.1*	*94.7*	*96.8*	*91.9*	*92.3*
2-2-222	78.4	*93.3*	*95.3*	*94.8*	*97.4*	*94.4*	*91*	*91.4*
2-2-218	77	*91.3*	*95.3*	*95*	*95.1*	*92.5*	*91.8*	*90.6*
2-2-217	78.4	*93.3*	*95.2*	*94.7*	*97.4*	*94.4*	*90.9*	*91.4*
2-2-213	78.4	*96.5*	*91.9*	*92.6*	*94.2*	*95.9*	*90.6*	*91*
2-2-212	77.8	*94*	*94.1*	*94.7*	*95.5*	*95*	*91.7*	*92.3*
2-2-211	77.7	*94.8*	*93.2*	*94*	*94.3*	*94.7*	*92.1*	*92.1*
2-2-111	78.6	*92.8*	*96*	*95.1*	*97.2*	*94.1*	*91.7*	*91.9*
2-2-109	79.2	*94.2*	*95.5*	*95*	*97.1*	*95.2*	*91.8*	*92.2*
2-2-107	75.4	80.7	80.9	80.2	81.8	81.4	79.8	80
2-2-102	79	*94.5*	*95.5*	*94.8*	*96.4*	*94.8*	*91.5*	*91.9*
W50	63.3	75.4	77.6	77.6	77.3	75.2	74.3	73.3
W36	64.3	76.3	79.2	78.8	78.9	76.3	75.8	74.8
W32	39.4	43.4	44.5	44.4	44.7	44.4	43.6	43.5
W31	65.9	78	78.9	78.9	79.9	77.3	76	76
W29	73.1	86.2	86.8	*87*	*87.3*	85.9	84.5	83.6
W25	37	41.7	41.5	41.3	42.3	42.8	40.6	40.6
W24	39.2	43.4	44.4	44.4	44.6	44.4	43.5	43.4
W10	38.7	42.9	44	43.9	44.1	43.9	42.8	42.7
W8	59	70.4	71.6	71.2	71.9	70.3	68.3	67.2
S43	65.6	80.2	78.3	79.3	78.9	80.7	77.3	76.3
S42	59.6	70.4	72.2	71.9	71.9	70.5	68.6	67.7
S40	35.4	40.3	40.6	40.7	40.8	40.4	39.8	39.6
S29	77.9	*92.3*	*96.3*	*95.6*	*95.9*	*93.5*	*92.3*	*90.9*
S26	38.2	43.1	42.4	42.3	43.4	43.1	41.1	41.3
S23	78.1	*91.6*	*96.8*	*95.7*	*96.5*	*93*	*92.3*	*90.7*
S17	78.3	*91.7*	*96.9*	*95.8*	*96.4*	*92.9*	*92.7*	*91.1*
S16	78.3	*91.7*	*96.9*	*95.8*	*96.4*	*92.9*	*92.7*	*91.1*
S6	75.6	*89.7*	*91.5*	*91*	*91.4*	*89.1*	*88.1*	86.9
S5	78.8	*93.3*	*96.2*	*95.4*	*95.9*	*93.6*	*92.6*	*91.5*
S4	78.4	*91.1*	*96.9*	*95.4*	*95.8*	*92.7*	*92*	*90.4*
S3	77.9	*90.6*	*96.6*	*95*	*95.3*	*92.2*	*91.4*	*89.8*

	2-2-223	2-2-222	2-2-218	2-2-217	2-2-213	2-2-212	2-2-211	2-2-111
6-2-510	94.1	97.3	94.2	97.3	93.9	95.1	93.7	96.7
6-2-508	93.9	96.6	94.8	96.6	93.6	95.8	94.3	96.9
6-2-507	97.4	94.7	92.7	94.7	96	95.3	96.2	94.2
6-2-504	92.2	89.5	88.9	89.5	92.3	90.2	90.9	89.2
6-2-502	93.4	96.1	95	96	92.8	95.5	94.3	96.9
6-2-501	43.7	43.5	43.7	43.5	43.3	43.5	43.3	43.7
6-2-415	93.8	96.5	94.1	96.5	93.1	94.6	93.7	96.2
6-2-409	93	92	90.2	92	91.9	91.2	91.4	91.9
6-2-406	97.2	94.3	93	94.3	95.3	95.2	94.7	94.1
6-2-405	94.1	95.9	96.2	95.9	93.5	96.5	93.8	96.6
6-2-401	93.8	94.2	95.3	94.2	92.9	93.7	94.8	95
6-2-310	92.7	92.9	91.4	92.9	91.7	92.1	92.6	93.2
6-2-309	93.5	93.6	91.8	93.6	92.6	92.3	93.3	93.9
6-2-306	96.5	95.2	93.8	95.2	94.9	94.4	93.7	95.1
6-2-304	96.8	94.6	92.8	94.6	95.5	95.8	95.5	94.4
6-2-303	95.3	94.9	92.9	94.9	94.1	94.2	94.7	94.4
6-2-210	96.8	95.4	94.1	95.4	95.6	94.9	94.4	95.5
6-2-207	87.3	86	85.7	86	86.4	85.6	85.2	86.2
6-2-206	95.3	95.5	93.7	95.5	94	97.1	94.7	95.3
6-2-203	97.2	93.8	92.4	93.7	95.5	94.9	94.6	93.5
6-2-120	96.2	96	93.7	96	94.8	96.5	95.4	95.8
6-2-119	93.6	95.6	97.2	95.6	93.5	95.4	93.2	96.1
6-2-118	94.1	96.8	94.8	96.8	94.1	96	94.8	97.3
6-2-117	93.3	96	94.3	95.9	92.9	95.2	94.6	96.8
6-2-113	93.7	95.7	97.3	95.7	93.4	95.5	93.3	96.2
6-2-112	93.1	95.8	95.6	95.7	92.5	95.2	94	96.5
6-2-111	93.1	95.8	95.6	95.7	92.5	95.2	94	96.5
6-2-110	93.4	95.6	97.2	95.6	92.6	95	93.5	96.5
6-2-108	92.6	95.4	94.7	95.4	92.1	94.2	92.8	95.7
6-2-106	94.4	95.7	94.3	95.7	94.1	94.5	93.1	95.8
6-2-105	94.4	95.7	94.3	95.7	94.1	94.5	93.1	95.8
6-2-104	94.4	96.9	94.6	96.9	93.9	95.2	94.1	96.7
6-2-103	95.1	96.5	94.9	96.5	94	97.3	95.1	97.1
2-2-508	95.1	96.1	94.4	96.3	94.1	96.9	95.4	96.2
2-2-503	94.2	95.8	94.1	95.8	93.1	94.7	93.9	95.8
2-2-501	93.6	95.5	94	95.5	93	95.4	93.8	96.4
2-2-418	93.9	96.8	94.5	96.8	93.5	95.5	94.2	96.8
2-2-402	93.5	96.7	94.4	96.7	93	95.4	93.7	96.9
2-2-330	83.3	82.9	83.6	82.8	82.6	82.3	83	83.4
2-2-329	93	92.6	90.9	92.6	92.4	92.4	93.3	92.6
2-2-324	78.2	78.4	77	78.4	78.4	77.8	77.7	78.6
2-2-320	96.2	93.3	91.3	93.3	96.5	94	94.8	92.8
2-2-318	92.3	95.3	95.3	95.2	91.9	94.1	93.2	96
2-2-317	94.1	94.8	95	94.7	92.6	94.7	94	95.1
2-2-309	94.7	97.4	95.1	97.4	94.2	95.5	94.3	97.2
2-2-306	96.8	94.4	92.5	94.4	95.9	95	94.7	94.1
2-2-305	91.9	91	91.8	90.9	90.6	91.7	92.1	91.7
2-2-304	92.3	91.4	90.6	91.4	91	92.3	92.1	91.9

	2-2-223	2-2-222	2-2-218	2-2-217	2-2-213	2-2-212	2-2-211	2-2-111
2-2-223	100	*94.6*	*92.6*	*94.6*	*96.3*	*95.4*	*95.6*	*94*
2-2-222	*94.6*	100	*94.7*	99.8	*93.9*	*95.3*	*94.1*	*97.1*
2-2-218	*92.6*	*94.7*	100	*94.6*	*91.8*	*94.6*	*92.8*	*95.8*
2-2-217	*94.6*	99.8	*94.6*	100	*93.9*	*95.3*	*94*	*97*
2-2-213	*96.3*	*93.9*	*91.8*	*93.9*	100	*94.5*	*94.2*	*93.7*
2-2-212	*95.4*	*95.3*	*94.6*	*95.3*	*94.5*	100	*94.9*	*95.8*
2-2-211	*95.6*	*94.1*	*92.8*	*94*	*94.2*	*94.9*	100	*94.8*
2-2-111	*94*	*97.1*	*95.8*	*97*	*93.7*	*95.8*	*94.8*	100
2-2-109	*94.9*	*96.1*	*95.3*	*96.1*	*94.7*	*95.9*	*94.6*	*97.6*
2-2-107	81.8	81.6	81.1	81.5	81	80.8	81.6	82.1
2-2-102	*95.4*	*96.5*	*94.5*	*96.5*	*94.6*	*97.1*	*95.8*	*96.6*
W50	75.2	77.2	76.8	77.2	75.7	76.7	75.6	77.5
W36	76	78.6	78.2	78.6	76.8	77.7	76.7	79.3
W32	44.1	44.3	44.7	44.3	43.7	44.8	44.6	45.2
W31	77.6	79.9	78.9	79.9	78.1	78.8	78.2	80
W29	85.8	86.6	86	86.6	86.4	85.9	85.5	*87*
W25	42.5	42.1	41.9	42.1	42.1	42	41.9	42.2
W24	44.1	44.4	44.6	44.4	43.7	44.7	44.5	45.1
W10	43.6	43.7	44.1	43.7	43.2	44	44	44.6
W8	69.9	71.5	70.1	71.5	70.8	70.6	69.7	70.9
S43	79.8	78.3	77.9	78.3	80.8	78.2	78.1	78.2
S42	69.9	71.4	71.4	71.4	70.4	71.5	70.5	72
S40	40.3	40.5	40.8	40.5	40.5	40.7	40.7	41.2
S29	*93.4*	*95.4*	*95*	*95.3*	*92.5*	*95.2*	*94.1*	*95.7*
S26	42.9	43.1	42.7	43.1	43.3	43	42.6	43.2
S23	*92.7*	*95.5*	*95.4*	*95.5*	*92.2*	*94.5*	*93.2*	*96.4*
S17	*92.8*	*95.6*	*95.7*	*95.5*	*92.1*	*94.6*	*93.1*	*96.2*
S16	*92.8*	*95.6*	*95.7*	*95.5*	*92.1*	*94.6*	*93.1*	*96.2*
S6	*88.9*	*90.6*	*90.6*	*90.6*	*89.9*	*90.7*	*89.1*	*91.2*
S5	*93.5*	*95.3*	*95.2*	*95.3*	*93.5*	*95.3*	*93.7*	*95.8*
S4	*92.4*	*95.3*	*95.4*	*95.2*	*91.8*	*94*	*93*	*95.7*
S3	*91.9*	*94.8*	*95*	*94.7*	*91.3*	*93.5*	*92.5*	*95.4*

	2-2-109	**2-2-107**	**2-2-102**	**W50**	**W36**	**W32**	**W31**	**W29**	**W25**	**W24**	**W10**
6-2-510	*96.6*	81.8	*96.4*	76.7	78.3	44.1	79.2	*87.4*	41.7	44	43.4
6-2-508	*97.2*	81.1	*97.2*	77.2	78.6	44.7	79.6	86.5	41.6	44.6	44
6-2-507	*95.6*	82.2	*95.9*	75.6	76.6	44.5	78.1	86.6	42.5	44.5	43.9
6-2-504	*90.3*	76.9	*90.7*	79.6	80	42.5	80.1	*90.1*	40.7	42.6	42.1
6-2-502	*96.4*	80.7	*96.4*	77.1	78.3	44.6	79.1	86.7	41.6	44.5	44.3
6-2-501	43.5	39.5	43.3	38	38.5	*92.7*	37.5	40	*89.7*	*92.9*	*93.5*
6-2-415	*96.1*	81.2	*95.3*	76.6	78.2	44.5	78.7	86.1	42.3	44.4	44
6-2-409	*93.3*	81.2	*91.8*	74.7	75.5	43.1	75.7	84.7	40.9	43.1	42.6
6-2-406	*95.1*	81.9	*94.6*	75.3	76.2	43.8	77.6	86	41.8	43.7	43.2
6-2-405	*96.4*	81.8	*95.9*	77.3	78.7	44.5	79.6	86.2	41.8	44.4	43.9
6-2-401	*95.1*	81.9	*94.5*	75.7	77.2	44.4	78.4	86.5	41.6	44.5	43.8
6-2-310	*93.9*	80.9	*92.3*	74	75.6	43.4	76.3	84.2	40.9	43.3	42.8
6-2-309	*94.8*	81.6	*93.1*	74.5	76	43.6	76.9	84.8	41.1	43.6	43
6-2-306	*95.9*	81.8	*94.1*	75.3	77	43.8	78	86.6	42.1	44	43.3
6-2-304	*95.4*	81.4	*95.4*	75.6	76.8	44.7	77.7	86	42.4	44.7	44.1
6-2-303	*95.4*	81.5	*94.4*	75.2	76.4	44.4	77.6	86.2	42	44.4	43.8
6-2-210	*96.4*	82.2	*94.6*	75.7	77.3	44.1	78.2	87	42.4	44.1	43.6
6-2-207	86.6	*89*	85.9	69.3	70.3	41.5	70.9	79.4	39.9	41.4	41
6-2-206	*95*	80.8	*96.5*	76.6	77.6	44.4	78.8	86.1	41.6	44.3	43.8
6-2-203	*94*	80.9	*94.5*	74.7	75.5	44	77.3	85.8	42.4	44	43.4
6-2-120	*95.8*	81.4	*97.2*	76.7	77.7	44.6	78.9	86	42.1	44.5	43.9
6-2-119	*96*	81.5	*95.6*	77.2	78.4	44.4	80	85.9	41.7	44.3	43.8
6-2-118	*97.2*	81.3	*97.3*	77.4	78.6	45	79.7	86.8	42	44.9	44.3
6-2-117	*96.1*	81.5	*96.3*	76.4	77.6	44.8	78.7	85.7	42	44.6	44.1
6-2-113	*96.1*	81.6	*95.7*	77.2	78.5	44.5	79.9	85.8	41.8	44.4	43.8
6-2-112	*96*	80.4	*96.1*	78.1	79.3	44.9	79	*87.2*	41.9	44.8	44.3
6-2-111	*96*	80.4	*96.1*	78.1	79.3	44.9	79	*87.2*	41.9	44.8	44.3
6-2-110	*96*	81.4	*95.5*	77	78.3	44.8	79.7	85.8	42.2	44.9	44.3
6-2-108	*95.4*	80.7	*95.4*	78.2	79.5	44.8	79.9	86.6	41.6	44.7	44.1
6-2-106	*97.2*	82	*94.8*	76.3	78	44.5	78.5	86.4	42.1	44.5	44.1
6-2-105	*97.2*	82	*94.8*	76.3	78	44.5	78.5	86.4	42.1	44.5	44.1
6-2-104	*96.8*	81.4	*96*	77.1	78.5	44.4	79.5	86.5	42.2	44.5	44
6-2-103	*96.6*	81.4	*97.4*	77.7	78.6	44.9	80	86.5	41.9	44.8	44.3
2-2-508	*95.9*	81	**99.4**	77.7	78.6	44.7	79.6	86	41.8	44.6	44.3
2-2-503	*95.8*	81.6	*95*	76	77.7	44.1	78.5	88.1	41.4	44.1	43.5
2-2-501	*96.1*	81.1	*96*	76.8	78.4	44.8	79.2	86.4	41.7	44.6	44.1
2-2-418	*96.4*	80.8	*96.9*	77.5	79	44.6	79.9	86.6	41.6	44.5	43.9
2-2-402	*96.2*	80.5	*96.6*	77.6	78.9	44.1	79.7	86.2	41.2	44	43.5
2-2-330	83.6	*94.4*	82.6	67.7	68.6	40.4	68.8	76.7	38.2	40.2	39.8
2-2-329	*93.9*	81.2	*92.9*	73.8	75.4	42.7	76.7	84.4	40.3	42.9	42.3
2-2-324	79.2	75.4	79	63.3	64.3	39.4	65.9	73.1	37	39.2	38.7
2-2-320	*94.2*	80.7	*94.5*	75.4	76.3	43.4	78	86.2	41.7	43.4	42.9
2-2-318	*95.5*	80.9	*95.5*	77.6	79.2	44.5	78.9	86.8	41.5	44.4	44
2-2-317	*95*	80.2	*94.8*	77.6	78.8	44.4	78.9	*87*	41.3	44.4	43.9
2-2-309	*97.1*	81.8	*96.4*	77.3	78.9	44.7	79.9	*87.3*	42.3	44.6	44.1
2-2-306	*95.2*	81.4	*94.8*	75.2	76.3	44.4	77.3	85.9	42.8	44.4	43.9
2-2-305	*91.8*	79.8	*91.5*	74.3	75.8	43.6	76	84.5	40.6	43.5	42.8
2-2-304	*92.2*	80	*91.9*	73.3	74.8	43.5	76	83.6	40.6	43.4	42.7



	2-2-109	**2-2-107**	**2-2-102**	**W50**	**W36**	**W32**	**W31**	**W29**	**W25**	**W24**	**W10**
2-2-223	*94.9*	81.8	*95.4*	75.2	76	44.1	77.6	85.8	42.5	44.1	43.6
2-2-222	*96.1*	81.6	*96.5*	77.2	78.6	44.3	79.9	86.6	42.1	44.4	43.7
2-2-218	*95.3*	81.1	*94.5*	76.8	78.2	44.7	78.9	86	41.9	44.6	44.1
2-2-217	*96.1*	81.5	*96.5*	77.2	78.6	44.3	79.9	86.6	42.1	44.4	43.7
2-2-213	*94.7*	81	*94.6*	75.7	76.8	43.7	78.1	86.4	42.1	43.7	43.2
2-2-212	*95.9*	80.8	*97.1*	76.7	77.7	44.8	78.8	85.9	42	44.7	44
2-2-211	*94.6*	81.6	*95.8*	75.6	76.7	44.6	78.2	85.5	41.9	44.5	44
2-2-111	*97.6*	82.1	*96.6*	77.5	79.3	45.2	80	*87*	42.2	45.1	44.6
2-2-109	**100**	82.2	*96.5*	77.1	78.9	45	79.6	*87.1*	42	45	44.4
2-2-107	82.2	**100**	81.3	64.5	65.5	40.2	66.9	73.8	38.2	40	39.6
2-2-102	*96.5*	81.3	**100**	78	78.9	44.6	80	86.4	41.7	44.5	44.1
W50	77.1	64.5	78	**100**	*88.9*	38.8	77.5	78.7	36.3	39	38.6
W36	78.9	65.5	78.9	*88.9*	**100**	39.4	78.2	79.5	36.8	39.6	39.1
W32	45	40.2	44.6	38.8	39.4	**100**	38.5	40.8	*90.4*	*97.1*	*95*
W31	79.6	66.9	80	77.5	78.2	38.5	**100**	81.1	38.8	38.6	38.2
W29	*87.1*	73.8	86.4	78.7	79.5	40.8	81.1	**100**	38.6	40.9	40.4
W25	42	38.2	41.7	36.3	36.8	*90.4*	38.8	38.6	**100**	*90.3*	*88.4*
W24	45	40	44.5	39	39.6	*97.1*	38.6	40.9	*90.3*	**100**	*95.1*
W10	44.4	39.6	44.1	38.6	39.1	*95*	38.2	40.4	*88.4*	*95.1*	**100**
W8	71	60.1	71.3	76.6	76.5	36.8	69	74	35.7	37	36.9
S43	79.1	68.5	78.4	81.9	80.8	39.7	71.4	77.3	38.5	39.9	39.8
S42	72	60.8	72.6	75.1	76.1	36.9	67	70.3	34.3	37	36.6
S40	41.2	35.8	40.7	39.9	41.1	80.5	35.9	38.4	75.3	81	79.6
S29	*95.9*	80.2	*95.9*	77.9	79.3	44.8	79.5	*87.3*	41.7	44.7	44.3
S26	43.1	38.5	42.9	40.5	40.4	86.7	40.7	43.9	81	86.3	85.1
S23	*96.2*	80.5	*95.4*	78.7	80.3	45	79.4	86.9	41.9	44.9	44.4
S17	*95.9*	80.7	*95.3*	78.6	80	44.6	79.3	86.9	41.5	44.5	44
S16	*95.9*	80.7	*95.3*	78.6	80	44.6	79.3	86.9	41.5	44.5	44
S6	*91.7*	75.9	*91.5*	81.1	81.5	42.6	82.9	*92.9*	39.9	42.6	42.2
S5	*96.4*	80.2	*96*	78.2	79.5	44.5	79.7	*88.7*	41.5	44.4	43.9
S4	*95.5*	81.4	*95.3*	78.3	79.5	44.8	79.2	86.5	41.5	44.5	44
S3	*95.2*	80.9	*94.8*	78.7	80	44.5	78.7	86	41.2	44.2	43.7

	W8	**S43**	**S42**	**S40**	**S29**	**S26**	**S23**	**S17**	**S16**	**S6**	**S5**	**S4**	**S3**
6-2-510	71.6	78.7	71.3	40.2	*95.5*	43	*95.4*	*95.4*	*95.4*	*91.1*	*95.7*	*95*	*94.6*
6-2-508	71.2	78.2	71.7	40.9	*96.3*	42.8	*95.6*	*95.6*	*95.6*	*92*	*96.6*	*95.5*	*95*
6-2-507	70.7	79.8	70.2	40.6	*94.1*	43.3	*93.1*	*93.2*	*93.2*	*90*	*94.4*	*93.1*	*92.5*
6-2-504	74.4	80.1	71.7	40.3	*90.4*	44.6	*90.1*	*90.4*	*90.4*	*93.8*	*91.1*	*89.5*	*89.5*
6-2-502	70.9	78.1	71.8	40.6	*96.7*	42.6	*96.1*	*95.9*	*95.9*	*91.9*	*96.6*	*95.7*	*95.3*
6-2-501	36.7	39.6	35.8	77.9	43.6	83.8	43.5	43.2	43.2	41.5	43.2	43.4	43.1
6-2-415	71.3	78.6	71.6	40.6	*94.8*	43.2	*95.3*	*95.1*	*95.1*	*89.9*	*94.5*	*94.6*	*94.1*
6-2-409	68.8	78.4	69.1	39.5	*91*	42.1	*91*	*91.5*	*91.5*	86.6	*91.2*	*91*	*90.5*
6-2-406	70	79.9	69.6	40.2	*93*	42.6	*92.9*	*92.9*	*92.9*	*89.1*	*93.6*	*92.6*	*92.1*
6-2-405	70.9	78.4	71.6	40.3	*94.9*	42.9	*95.4*	*95.8*	*95.8*	*90.7*	*95.3*	*95.3*	*94.9*
6-2-401	69.5	77.6	70.3	40.4	*93.5*	42.5	*93.5*	*93.7*	*93.7*	*89.8*	*94.4*	*93.6*	*93*
6-2-310	67.6	76.9	68.4	39.2	*91.6*	41.5	*91.6*	*91.6*	*91.6*	*87.5*	*92.2*	*91.7*	*91.3*
6-2-309	68.1	77.4	68.8	39.6	*92.1*	41.7	*92.1*	*92.1*	*92.1*	*88.2*	*92.8*	*92.1*	*91.8*
6-2-306	70	79.5	70.4	40.2	*93.8*	42.5	*94.1*	*94.1*	*94.1*	*89.8*	*94.4*	*93.6*	*93.1*
6-2-304	70.5	79.6	70.4	40.7	*94.1*	43.5	*93.3*	*93.4*	*93.4*	*89.5*	*94.1*	*93*	*92.5*
6-2-303	70.5	79	69.9	40.7	*94.2*	43.2	*93.6*	*93.6*	*93.6*	*90*	*94.7*	*93.3*	*92.9*
6-2-210	70.2	80.3	70.3	40.4	*94*	42.8	*94.3*	*94.4*	*94.4*	*89.9*	*94.5*	*93.8*	*93.3*
6-2-207	64.2	74.3	65.9	37.4	84.7	40.4	84.6	85.2	85.2	81.2	85.6	85.2	84.7
6-2-206	71.1	78.2	71.4	40.4	*94.9*	42.9	*94.1*	*94.3*	*94.3*	*90.2*	*94.8*	*93.7*	*93.4*
6-2-203	69.6	79	69.7	40	*92.9*	42.6	*92.5*	*92.4*	*92.4*	*88.9*	*93.3*	*91.9*	*91.4*
6-2-120	71	78.4	71.3	40.5	*95.3*	43.2	*94.5*	*94.4*	*94.4*	*90.4*	*95.1*	*94.3*	*93.8*
6-2-119	70.3	78.2	71.4	40.7	*93.9*	43	*94.5*	*94.9*	*94.9*	*90.6*	*95.1*	*94.3*	*93.9*
6-2-118	71.3	78.1	72.1	40.9	*96.5*	43.2	*95.9*	*95.7*	*95.7*	*92.3*	*97*	*95.5*	*95.1*
6-2-117	70.4	77.4	71.1	40.7	*95.9*	42.7	*95.3*	*95.1*	*95.1*	*91.1*	*95.7*	*94.9*	*94.4*
6-2-113	70.2	78.2	71.5	40.7	*94*	42.9	*94.6*	*95*	*95*	*90.5*	*95.1*	*94.4*	*94*
6-2-112	72.1	79	72.9	41.4	*97.5*	42.8	*97.3*	*97.1*	*97.1*	*92.8*	*97.2*	*96.7*	*96.5*
6-2-111	72.1	79	72.9	41.4	*97.5*	42.8	*97.3*	*97.1*	*97.1*	*92.8*	*97.2*	*96.7*	*96.5*
6-2-110	70.1	77.8	71.4	40.9	*94.4*	43	*95*	*95*	*95*	*90.2*	*94.7*	*94.9*	*94.5*
6-2-108	71.1	78.2	72.6	40.8	*95.5*	43	*96.2*	*96.7*	*96.7*	*91.1*	*95.3*	*96.9*	*96.5*
6-2-106	71	79.2	70.7	40.7	*94.3*	43.4	*94.8*	*94.9*	*94.9*	*90.1*	*94.7*	*94.2*	*93.8*
6-2-105	71	79.2	70.7	40.7	*94.3*	43.4	*94.8*	*94.9*	*94.9*	*90.1*	*94.7*	*94.2*	*93.8*
6-2-104	71.2	78.7	71.6	41	*95.5*	43.2	*95.9*	*95.7*	*95.7*	*90.9*	*95.4*	*95.3*	*94.8*
6-2-103	70.6	78.3	72.1	41	*95.8*	43.1	*95.5*	*95.6*	*95.6*	*91.3*	*95.9*	*94.9*	*94.4*
2-2-508	71.1	78.1	72.3	40.8	*95.8*	43	*95.2*	*95.1*	*95.1*	*91.2*	*95.6*	*95.1*	*94.6*
2-2-503	70.6	78.4	70.7	40.2	*94.5*	42.6	*94.3*	*94.3*	*94.3*	*90.5*	*95*	*93.9*	*93.4*
2-2-501	71.2	77.6	71.4	40.8	*95.5*	42.9	*95.1*	*95.3*	*95.3*	*91.1*	*95.4*	*94.5*	*94.1*
2-2-418	70.9	78.2	71.5	40.8	*95.5*	42.8	*95.2*	*95.3*	*95.3*	*91.1*	*95.7*	*95*	*94.5*
2-2-402	70.9	78.3	71.8	40.4	*95.1*	42.5	*95.3*	*95.5*	*95.5*	*91*	*95.6*	*95*	*94.4*
2-2-330	62.6	72	63.7	36.4	83.3	38.7	84	84.2	84.2	79	83.2	84.5	83.9
2-2-329	67.6	77.2	69.3	39.2	*91.7*	40.8	*91.9*	*91.5*	*91.5*	*87.5*	*92.1*	*91.3*	*90.7*
2-2-324	59	65.6	59.6	35.4	77.9	38.2	78.1	78.3	78.3	75.6	78.8	78.4	77.9
2-2-320	70.4	80.2	70.4	40.3	*92.3*	43.1	*91.6*	*91.7*	*91.7*	*89.7*	*93.3*	*91.1*	*90.6*
2-2-318	71.6	78.3	72.2	40.6	*96.3*	42.4	*96.8*	*96.9*	*96.9*	*91.5*	*96.2*	*96.9*	*96.6*
2-2-317	71.2	79.3	71.9	40.7	*95.6*	42.3	*95.7*	*95.8*	*95.8*	*91*	*95.4*	*95.4*	*95*
2-2-309	71.9	78.9	71.9	40.8	*95.9*	43.4	*96.5*	*96.4*	*96.4*	*91.4*	*95.9*	*95.8*	*95.3*
2-2-306	70.3	80.7	70.5	40.4	*93.5*	43.1	*93*	*92.9*	*92.9*	*89.1*	*93.6*	*92.7*	*92.2*
2-2-305	68.3	77.3	68.6	39.8	*92.3*	41.1	*92.3*	*92.7*	*92.7*	*88.1*	*92.6*	*92*	*91.4*
2-2-304	67.2	76.3	67.7	39.6	*90.9*	41.3	*90.7*	*91.1*	*91.1*	*86.9*	*91.5*	*90.4*	*89.8*

	W8	S43	S42	S40	S29	S26	S23	S17	S16	S6	S5	S4	S3
2-2-223	69.9	79.8	69.9	40.3	*93.4*	42.9	*92.7*	*92.8*	*92.8*	*88.9*	*93.5*	*92.4*	*91.9*
2-2-222	71.5	78.3	71.4	40.5	*95.4*	43.1	*95.5*	*95.6*	*95.6*	*90.6*	*95.3*	*95.3*	*94.8*
2-2-218	70.1	77.9	71.4	40.8	*95*	42.7	*95.4*	*95.7*	*95.7*	*90.6*	*95.2*	*95.4*	*95*
2-2-217	71.5	78.3	71.4	40.5	*95.3*	43.1	*95.5*	*95.5*	*95.5*	*90.6*	*95.3*	*95.2*	*94.7*
2-2-213	70.8	80.8	70.4	40.5	*92.5*	43.3	*92.2*	*92.1*	*92.1*	*89.9*	*93.5*	*91.8*	*91.3*
2-2-212	70.6	78.2	71.5	40.7	*95.2*	43	*94.5*	*94.6*	*94.6*	*90.7*	*95.3*	*94*	*93.5*
2-2-211	69.7	78.1	70.5	40.7	*94.1*	42.6	*93.2*	*93.1*	*93.1*	*89.1*	*93.7*	*93*	*92.5*
2-2-111	70.9	78.2	72	41.2	*95.7*	43.2	*96.4*	*96.2*	*96.2*	*91.2*	*95.8*	*95.7*	*95.4*
2-2-109	71	79.1	72	41.2	*95.9*	43.1	*96.2*	*95.9*	*95.9*	*91.7*	*96.4*	*95.5*	*95.2*
2-2-107	60.1	68.5	60.8	35.8	80.2	38.5	80.5	80.7	80.7	75.9	80.2	81.4	80.9
2-2-102	71.3	78.4	72.6	40.7	*95.9*	42.9	*95.4*	*95.3*	*95.3*	*91.5*	*96*	*95.3*	*94.8*
W50	76.6	81.9	75.1	39.9	77.9	40.5	78.7	78.6	78.6	81.1	78.2	78.3	78.7
W36	76.5	80.8	76.1	41.1	79.3	40.4	80.3	80	80	81.5	79.5	79.5	80
W32	36.8	39.7	36.9	80.5	44.8	86.7	45	44.6	44.6	42.6	44.5	44.8	44.5
W31	69	71.4	67	35.9	79.5	40.7	79.4	79.3	79.3	82.9	79.7	79.2	78.7
W29	74	77.3	70.3	38.4	*87.3*	43.9	86.9	86.9	86.9	92.9	*88.7*	86.5	86
W25	35.7	38.5	34.3	75.3	41.7	81	41.9	41.5	41.5	39.9	41.5	41.5	41.2
W24	37	39.9	37	81	44.7	86.3	44.9	44.5	44.5	42.6	44.4	44.5	44.2
W10	36.9	39.8	36.6	79.6	44.3	85.1	44.4	44	44	42.2	43.9	44	43.7
W8	**100**	75.3	69.8	38.7	72	40.4	72.1	72	72	75.5	72	71.5	71.9
S43	75.3	**100**	77.6	41	78.7	39.9	79.8	79.2	79.2	78	79.4	78.7	79.1
S42	69.8	77.6	**100**	39.2	73.1	36.7	73.9	73.6	73.6	72.6	72.7	73	73.4
S40	38.7	41	39.2	**100**	41	76.7	41.7	41.3	41.3	39.9	41.1	40.9	41.1
S29	72	78.7	73.1	41	**100**	42.7	*96.9*	*96.5*	*96.5*	*93*	*97.5*	*96.5*	*95.9*
S26	40.4	39.9	36.7	76.7	42.7	**100**	42.8	42.6	42.6	44.8	43	42.6	42.4
S23	72.1	79.8	73.9	41.7	*96.9*	42.8	**100**	*98.2*	*98.2*	*91.8*	*96.3*	*97.2*	*97.6*
S17	72	79.2	73.6	41.3	*96.5*	42.6	*98.2*	**100**	**100**	*91.7*	*96.2*	*97.4*	*97.4*
S16	72	79.2	73.6	41.3	*96.5*	42.6	*98.2*	**100**	**100**	*91.7*	*96.2*	*97.4*	*97.4*
S6	75.5	78	72.6	39.9	*93*	44.8	*91.8*	*91.7*	*91.7*	**100**	*95.3*	*91.6*	*91*
S5	72	79.4	72.7	41.1	*97.5*	43	*96.3*	*96.2*	*96.2*	*95.3*	**100**	*96*	*95.4*
S4	71.5	78.7	73	40.9	*96.5*	42.6	*97.2*	*97.4*	*97.4*	*91.6*	*96*	**100**	**99.4**
S3	71.9	79.1	73.4	41.1	*95.9*	42.4	*97.6*	*97.4*	*97.4*	*91*	*95.4*	**99.4**	**100**

Table A4: Complete similarity matrix for summer and winter clones that were most closely related to *Methanoregula boonei*, *Methanospirillum hungatei* and *Methanosphaerula palustris* compared to Grosser's clones that were most closely related to *Methanospirillum*. Italicized numbers indicate a similarity index of 87 or greater; bolded numbers indicate sequences with a similarity index of 98.7 or greater.

	W44	W41	W38	W35	W28	W15	W12	S49	S45	S44	S41	S39
W44	**100**	36.3	63.5	79	85.3	36	67.6	63.1	37.7	80.7	38	75
W41	36.3	**100**	32.6	34.8	35.6	76.1	30.5	26.6	77.6	36.9	80	37.5
W38	63.5	32.6	**100**	70.4	63.2	36.1	47.3	52.5	34.6	73.9	35	66.9
W35	79	34.8	70.4	**100**	78.9	38.3	62.7	63.5	36.2	*88.3*	37.1	74.8
W28	85.3	35.6	63.2	78.9	**100**	36.1	63.1	60.8	37.2	80.1	39.1	73.8
W15	36	76.1	36.1	38.3	36.1	**100**	33.2	29.3	78.5	40.9	83.9	38.4
W12	67.6	30.5	47.3	62.7	63.1	33.2	**100**	49.1	31	62.3	32.7	56.9
S49	63.1	26.6	52.5	63.5	60.8	29.3	49.1	**100**	27.4	65.3	28.3	55.1
S45	37.7	77.6	34.6	36.2	37.2	78.5	31	27.4	100	38.7	81.6	39.2
S44	80.7	36.9	73.9	*88.3*	80.1	40.9	62.3	65.3	38.7	**100**	39.6	79.5
S41	38	80	35	37.1	39.1	83.9	32.7	28.3	81.6	39.6	**100**	39.5
S39	75	37.5	66.9	74.8	73.8	38.4	56.9	55.1	39.2	79.5	39.5	**100**
S36	79.9	35.9	73.8	*88.5*	80.2	40	61.6	64.5	37.9	*93.1*	39	78.5
S35	52.3	27.6	55.1	56.7	52.3	30.5	33.1	44.3	28.8	61.7	28.3	53.1
S34	38.5	72.6	35	39.1	38.1	85.4	34.4	30.2	79.1	40.2	79.3	38.2
S32	76.3	37	73.3	83.7	77.2	40.7	57.3	63	38.3	*89.5*	38.8	77.4
S31	73.7	35.6	80.5	81.5	73	39.5	56.2	59.4	38	86.2	38.5	77.5
S28	36.3	73	36.9	38.1	36.2	*87.5*	32.9	29.1	81	40.9	79.9	39.4
S27	66.2	32.3	49.2	60.5	60.6	33.7	75.5	47.7	32.5	60.1	33	58.8
S24	38.7	72.8	35.9	39.2	38.1	81.1	34.2	30.2	83.3	40.4	76.1	39
S22	*88.2*	35.4	70.3	86.4	83.2	38.4	68.9	69.3	36.4	*88.2*	37.4	75.4
S21	37.9	75.5	36.2	39.9	37.7	*88.7*	34.5	30.4	77.8	40.7	82.6	38.4
S20	38.9	75.3	36.1	39.8	38.6	*88.3*	35.3	30.9	77.5	40.9	81.3	38.6
S19	38.6	76.2	36.2	39.4	37.9	85.2	35	30.7	76.7	40.6	79.4	39.3
S18	38.5	76.1	36.2	39.4	37.8	85.2	34.9	30.7	76.6	40.6	79.3	39.3
S15	39	72.9	36.2	39.4	38.4	81.4	34.6	30.3	83.7	40.6	76.5	39.4
S14	38.6	73	36.1	39.2	38	81.2	34.2	30.1	83.7	40.3	76.5	39.1
S13	38.3	72.5	36.3	39.5	37.8	81.8	33.9	30.2	83.4	40.7	76	39.1
S12	38.1	72.6	36.3	39.5	37.8	82	33.8	30	83.5	40.8	76.2	39.1
S11	35.6	67.7	36.3	36.5	35.2	79.8	32.2	29.1	69.9	37.5	73.2	35.4
S10	34.9	67	35.8	36	34.5	79.6	32.1	28.6	69	36.8	72.4	34.8
S9	40.6	75.9	34.5	38.9	39.9	81	35.1	30	82.7	39.8	81.3	38.9
S8	38.4	77.5	36	39.7	38.4	84.8	35.4	31	77.3	40.7	79.7	38.7
S7	37.9	77.2	35.8	39.4	37.8	84.5	34.9	30.6	76.8	40.3	79.4	38.3
S2	39.2	73.1	35.1	39.8	38.5	82.1	34.6	30.6	81.8	40.8	77.3	38.6
S1	38.9	72.7	32	37.1	39.6	72.5	33.9	28.2	79.5	37.8	77.5	36.3
06-02-503	38.1	74.3	36.3	39.5	37.9	*87.3*	34.8	30.8	76.5	40.6	81.2	38.1
06-02-204	38.3	73.9	36.4	40	38.3	*87.9*	34.8	31.2	76.6	41.1	81.5	38.2
06-02-121	37.9	73.3	35.5	39.6	37.8	86.3	34.7	30.6	75.6	40.3	80.2	37.8
02-02-507	38.2	74.9	36.5	39.7	38.1	*87.3*	34.8	30.9	76.5	40.8	81	38.4
02-02-506	38	74.6	36.5	39.2	37.6	85.4	34.7	30.6	77	40.3	79	38.4
02-02-505	38.6	75.2	36.7	39.9	38.2	*87.7*	35.1	31.1	76.8	40.9	81.2	38.5
02-02-504	38.6	75	36.8	40	38.2	*87*	34.9	31	77.3	40.9	80.4	39.3
02-02-412	38.5	74.7	36.6	39.9	38.1	86.5	34.7	31	76.7	41.1	80.8	38.5
02-02-404	37.9	74	36.6	39.5	37.6	*87.5*	34.9	30.4	75.6	40.6	79.6	37.6
02-02-401	38.4	74.5	36.3	39.4	37.8	86.8	34.7	30.7	75.8	40.5	79.9	37.8
02-02-319	37.8	72.8	35.4	39.1	37.7	83.3	34.9	30.3	74	39.8	77.3	37.8

	S36	**S35**	**S34**	**S32**	**S31**	**S28**	**S27**	**S24**	**S22**	**S21**	**S20**	**S19**
W44	79.9	52.3	38.5	76.3	73.7	36.3	66.2	38.7	*88.2*	37.9	38.9	38.6
W41	35.9	27.6	72.6	37	35.6	73	32.3	72.8	35.4	75.5	75.3	76.2
W38	73.8	55.1	35	73.3	80.5	36.9	49.2	35.9	70.3	36.2	36.1	36.2
W35	*88.5*	56.7	39.1	83.7	81.5	38.1	60.5	39.2	86.4	39.9	39.8	39.4
W28	80.2	52.3	38.1	77.2	73	36.2	60.6	38.1	83.2	37.7	38.6	37.9
W15	40	30.5	85.4	40.7	39.5	*87.5*	33.7	81.1	38.4	*88.7*	*88.3*	85.2
W12	61.6	33.1	34.4	57.3	56.2	32.9	75.5	34.2	68.9	34.5	35.3	35
S49	64.5	44.3	30.2	63	59.4	29.1	47.7	30.2	69.3	30.4	30.9	30.7
S45	37.9	28.8	79.1	38.3	38	81	32.5	83.3	36.4	77.8	77.5	76.7
S44	*93.1*	61.7	40.2	*89.5*	86.2	40.9	60.1	40.4	*88.2*	40.7	40.9	40.6
S41	39	28.3	79.3	38.8	38.5	79.9	33	76.1	37.4	82.6	81.3	79.4
S39	78.5	53.1	38.2	77.4	77.5	39.4	58.8	39	75.4	38.4	38.6	39.3
S36	**100**	61	40.1	*88*	85.2	40.4	59.7	40.2	*87.5*	40.5	40.7	40.3
S35	61	**100**	29	65.5	60.3	31.3	34	28.8	60.8	29.9	29.7	29.3
S34	40.1	29	**100**	38.4	38.1	82.1	35.7	*93.4*	39.9	*92.1*	*92.2*	*90.8*
S32	88	65.5	38.4	**100**	85.8	41.2	56.3	38.6	85.2	39.5	39.8	39.4
S31	85.2	60.3	38.1	85.8	**100**	41.2	56.6	39.2	80.6	38.8	38.8	39.3
S28	40.4	31.3	82.1	41.2	41.2	**100**	34	82.4	38.6	84.3	83.9	81.8
S27	59.7	34	35.7	56.3	56.6	34	**100**	35.8	67.3	35.6	36	36.8
S24	40.2	28.8	*93.4*	38.6	39.2	82.4	35.8	**100**	40.4	*89.1*	*90.7*	*90.6*
S22	*87.5*	60.8	39.9	85.2	80.6	38.6	67.3	40.4	**100**	40.8	41.3	41.2
S21	40.5	29.9	*92.1*	39.5	38.8	84.3	35.6	*89.1*	40.8	**100**	*95.1*	*93.5*
S20	40.7	29.7	*92.2*	39.8	38.8	83.9	36	*90.7*	41.3	*95.1*	**100**	*94*
S19	40.3	29.3	*90.8*	39.4	39.3	81.8	36.8	*90.6*	41.2	*93.5*	*94*	**100**
S18	40.3	29.3	*90.8*	39.4	39.3	81.8	36.8	*90.5*	41.1	*93.5*	*93.9*	**99.90**
S15	40.5	29.1	*93.6*	38.8	39.4	82.7	36.2	**99**	40.7	*89.5*	*91*	*90.8*
S14	40.1	28.7	*93.5*	38.5	39.3	82.5	35.8	**98.90**	40.3	*89.4*	*90.7*	*90.5*
S13	40.5	29.3	*93*	38.9	39.5	83.1	35.5	*97.6*	40.1	*89.7*	*89.4*	*89.2*
S12	40.6	29.3	*92.8*	38.9	39.5	83.3	35.4	*97.4*	39.9	*89.9*	*89.2*	*89*
S11	37.2	27.1	83.3	36.4	35.6	75.8	33.5	81.2	38.1	86.7	86.9	85
S10	36.6	26.8	82.6	35.7	35.2	74.6	32.7	80.2	37.5	86.5	86.5	84
S9	39.8	28	*92*	38.5	37.9	78.3	35.9	*89.9*	39.8	*87.5*	*88.7*	*87.7*
S8	40.6	29.4	*91.4*	39.7	39.1	82.7	36.2	*91.1*	41.2	*92.7*	*94.7*	*95.2*
S7	40.2	29.4	*90.6*	39.4	38.8	82.5	35.7	*90.2*	40.6	*91.8*	*93.8*	*94.2*
S2	40.3	28.9	*94.5*	39	38.6	80.6	35.7	*96.6*	40.6	*89.8*	*91.2*	*90.2*
S1	37.4	25.4	83.1	35.9	35.6	71	34.6	85.2	37.7	80	81.4	80.3
06-02-503	40.3	29.8	*91.9*	39.4	38.6	82.9	36	*88.8*	41.1	*97.2*	*94.8*	*93.2*
06-02-204	40.8	29.7	*92.5*	39.9	38.8	83.4	35.6	*89.3*	40.9	*95.5*	*95.2*	*92.5*
06-02-121	40.1	30	*92*	39.1	38.7	82.7	35.6	*88.6*	40.9	*95.2*	*93.5*	*92*
02-02-507	40.6	30	*91.3*	39.7	38.9	83.1	36.1	*88.8*	41.2	*96.2*	*95*	*93.5*
02-02-506	40	29.1	*90.7*	39	39.1	83.3	36.5	*89.6*	40.3	*93.4*	*93.5*	*94.1*
02-02-505	40.7	30.2	*91.5*	39.7	39	83.4	36.4	*89*	41.5	*96.5*	*95.2*	*93.6*
02-02-504	40.6	29.7	*91.6*	39.7	39.4	84.5	36.5	*89.8*	41.1	*95.3*	*94.6*	*95.1*
02-02-412	40.7	30.2	*92*	39.7	39	82.7	35.8	*89.5*	41.4	*95.9*	*95.3*	*93.3*
02-02-404	40.2	29.4	*89.4*	39.5	39	81.8	35.6	86.8	40.2	*93.8*	*94.4*	*90.7*
02-02-401	40.1	29.6	*91*	39.3	38.6	82.3	35.5	88.3	40.5	*93.9*	*95.6*	*91.9*
02-02-319	39.7	28.8	*89.3*	38.7	38.1	78.4	36.4	86.8	40.2	*91.5*	*90.5*	*90.6*

	S18	**S15**	**S14**	**S13**	**S12**	**S11**	**S10**	**S9**	**S8**	**S7**	**S2**	**S1**
W44	38.5	39	38.6	38.3	38.1	35.6	34.9	40.6	38.4	37.9	39.2	38.9
W41	76.1	72.9	73	72.5	72.6	67.7	67	75.9	77.5	77.2	73.1	72.7
W38	36.2	36.2	36.1	36.3	36.3	36.3	35.8	34.5	36	35.8	35.1	32
W35	39.4	39.4	39.2	39.5	39.5	36.5	36	38.9	39.7	39.4	39.8	37.1
W28	37.8	38.4	38	37.8	37.8	35.2	34.5	39.9	38.4	37.8	38.5	39.6
W15	85.2	81.4	81.2	81.8	82	79.8	79.6	81	84.8	84.5	82.1	72.5
W12	34.9	34.6	34.2	33.9	33.8	32.2	32.1	35.1	35.4	34.9	34.6	33.9
S49	30.7	30.3	30.1	30.2	30	29.1	28.6	30	31	30.6	30.6	28.2
S45	76.6	83.7	83.7	83.4	83.5	69.9	69	82.7	77.3	76.8	81.8	79.5
S44	40.6	40.6	40.3	40.7	40.8	37.5	36.8	39.8	40.7	40.3	40.8	37.8
S41	79.3	76.5	76.5	76	76.2	73.2	72.4	81.3	79.7	79.4	77.3	77.5
S39	39.3	39.4	39.1	39.1	39.1	35.4	34.8	38.9	38.7	38.3	38.6	36.3
S36	40.3	40.5	40.1	40.5	40.6	37.2	36.6	39.8	40.6	40.2	40.3	37.4
S35	29.3	29.1	28.7	29.3	29.3	27.1	26.8	28	29.4	29.4	28.9	25.4
S34	*90.8*	*93.6*	*93.5*	*93*	*92.8*	83.3	82.6	*92*	*91.4*	*90.6*	*94.5*	83.1
S32	39.4	38.8	38.5	38.9	38.9	36.4	35.7	38.5	39.7	39.4	39	35.9
S31	39.3	39.4	39.3	39.5	39.5	35.6	35.2	37.9	39.1	38.8	38.6	35.6
S28	81.8	82.7	82.5	83.1	83.3	75.8	74.6	78.3	82.7	82.5	80.6	71
S27	36.8	36.2	35.8	35.5	35.4	33.5	32.7	35.9	36.2	35.7	35.7	34.6
S24	*90.5*	**99**	**98.9**	*97.6*	*97.4*	81.2	80.2	*89.9*	*91.1*	*90.2*	*96.6*	85.2
S22	41.1	40.7	40.3	40.1	39.9	38.1	37.5	39.8	41.2	40.6	40.6	37.7
S21	*93.5*	*89.5*	*89.4*	*89.7*	*89.9*	86.7	86.5	87.5	*92.7*	*91.8*	*89.8*	80
S20	*93.9*	*91*	*90.7*	*89.4*	*89.2*	86.9	86.5	*88.7*	*94.7*	*93.8*	*91.2*	81.4
S19	**99.9**	*90.8*	*90.5*	*89.2*	*89*	85	84	*87.7*	*95.2*	*94.2*	*90.2*	80.3
S18	**100**	*90.7*	*90.4*	*89.2*	*89*	85	84	*87.6*	*95.1*	*94.1*	*90.1*	80.2
S15	*90.7*	**100**	99.1	*97.8*	*97.6*	81.5	80.6	*90.1*	*91.2*	*90.4*	*96.8*	85.5
S14	*90.4*	**99.1**	**100**	*97.7*	*97.5*	81.4	80.5	*90.3*	*91.1*	*90.4*	*96.9*	85.6
S13	*89.2*	*97.8*	*97.7*	**100**	**99.8**	80.6	79.7	*88.8*	*89.7*	*88.8*	*95.4*	84.1
S12	*89*	*97.6*	*97.5*	**99.8**	**100**	80.5	79.5	*88.6*	*89.5*	*88.6*	*95.2*	83.9
S11	85	81.5	81.4	80.6	80.5	**100**	*97.1*	79.8	84.6	83.8	81.9	72.9
S10	84	80.6	80.5	79.7	79.5	*97.1*	**100**	78.9	83.8	82.9	81.1	72.1
S9	*87.6*	*90.1*	*90.3*	*88.8*	*88.6*	79.8	78.9	**100**	*88.1*	*87.1*	*91.2*	86.3
S8	*95.1*	*91.2*	*91.1*	*89.7*	*89.5*	84.6	83.8	*88.1*	**100**	**98.8**	*91.2*	81.4
S7	*94.1*	*90.4*	*90.4*	*88.8*	*88.6*	83.8	82.9	*87.1*	**98.8**	**100**	*90.2*	80.4
S2	*90.1*	*96.8*	*96.9*	*95.4*	*95.2*	81.9	81.1	*91.2*	*91.2*	*90.2*	**100**	*88.3*
S1	80.2	85.5	85.6	84.1	83.9	72.9	72.1	86.3	*81.4*	80.4	*88.3*	**100**
06-02-503	*93.3*	*89.2*	*89.1*	*88.6*	*88.4*	*87.5*	*87.3*	*87.3*	*92.4*	*91.4*	*89.5*	79.8
06-02-204	*92.5*	*89.5*	*89.4*	*89*	*88.8*	*87.2*	86.1	*87.8*	*92.4*	*91.5*	*89.9*	80.2
06-02-121	*92.1*	*88.8*	*88.7*	*88.3*	*88.1*	*85.7*	85.8	*88.2*	*91.6*	*91*	*89.3*	79.5
02-02-507	*93.5*	*89.2*	*89.1*	*88.6*	*88.4*	*87.7*	87.3	*87.6*	*93.3*	*92.3*	*89.7*	79.9
02-02-506	*94.1*	*90*	*89.9*	*89.6*	*89.4*	86.4	84.9	86.9	*93.7*	*92.8*	*88.8*	79.4
02-02-505	*93.7*	*89.4*	*89.3*	*88.8*	*88.6*	*88*	*87.6*	*87.8*	*93.5*	*92.6*	*89.8*	80
02-02-504	*95.2*	*90.1*	*90*	*89.5*	*89.3*	*87.1*	86.2	*87.2*	*93.2*	*92.2*	*89.6*	79.7
02-02-412	*93.3*	*89.9*	*89.6*	*89.5*	*89.3*	86.9	86.7	*87.7*	*93.3*	*92.3*	*90.2*	80.2
02-02-404	*90.8*	*87.1*	*87*	*87.3*	*87.3*	85.6	85.7	85.4	*91*	*90*	*88*	78.8
02-02-401	*92*	*88.5*	*88.3*	*87.8*	*87.7*	86.9	86.2	*87.1*	*92.5*	*91.6*	*89.3*	80
02-02-319	*90.6*	*87*	86.7	86.5	86.3	83.4	83.3	85.3	*89.9*	*89*	*87.8*	78.1

	06-02-503	06-02-204	06-02-121	02-02-507	02-02-506
W44	38.1	38.3	37.9	38.2	38
W41	74.3	73.9	73.3	74.9	74.6
W38	36.3	36.4	35.5	36.5	36.5
W35	39.5	40	39.6	39.7	39.2
W28	37.9	38.3	37.8	38.1	37.6
W15	*87.3*	*87.9*	86.3	*87.3*	85.4
W12	34.8	34.8	34.7	34.8	34.7
S49	30.8	31.2	30.6	30.9	30.6
S45	76.5	76.6	75.6	76.5	77
S44	40.6	41.1	40.3	40.8	40.3
S41	81.2	81.5	80.2	81	79
S39	38.1	38.2	37.8	38.4	38.4
S36	40.3	40.8	40.1	40.6	40
S35	29.8	29.7	30	30	29.1
S34	*91.9*	*92.5*	*92*	*91.3*	*90.7*
S32	39.4	39.9	39.1	39.7	39
S31	38.6	38.8	38.7	38.9	39.1
S28	82.9	83.4	82.7	83.1	83.3
S27	36	35.6	35.6	36.1	36.5
S24	*88.8*	*89.3*	*88.6*	*88.8*	*89.6*
S22	41.1	40.9	40.9	41.2	40.3
S21	*97.2*	*95.5*	*95.2*	*96.2*	*93.4*
S20	*94.8*	*95.2*	*93.5*	*95*	*93.5*
S19	*93.2*	*92.5*	*92*	*93.5*	*94.1*
S18	*93.3*	*92.5*	*92.1*	*93.5*	*94.1*
S15	*89.2*	*89.5*	*88.8*	*89.2*	*90*
S14	*89.1*	*89.4*	*88.7*	*89.1*	*89.9*
S13	*88.6*	*89*	*88.3*	*88.6*	*89.6*
S12	*88.4*	*88.8*	*88.1*	*88.4*	*89.4*
S11	*87.5*	*87.2*	85.7	*87.7*	86.4
S10	*87.3*	86.1	85.8	*87.3*	84.9
S9	*87.3*	*87.8*	*88.2*	*87.6*	86.9
S8	*92.4*	*92.4*	*91.6*	*93.3*	*93.7*
S7	*91.4*	*91.5*	*91*	*92.3*	*92.8*
S2	*89.5*	*89.9*	*89.3*	*89.7*	*88.8*
S1	79.8	80.2	79.5	79.9	79.4
06-02-503	**100**	*96.8*	*96.7*	*97.6*	*94.2*
06-02-204	*96.8*	**100**	*95.1*	*96.3*	*94.3*
06-02-121	*96.7*	*95.1*	**100**	*95.6*	*93.4*
02-02-507	*97.6*	*96.3*	*95.6*	**100**	*94.8*
02-02-506	*94.2*	*94.3*	*93.4*	*94.8*	**100**
02-02-505	*97.9*	*96.6*	*95.9*	**99.1**	*95.1*
02-02-504	*96.3*	*95.9*	*94.7*	*96.3*	*96.5*
02-02-412	*97.3*	*95.8*	*95.8*	*97.3*	*94.1*
02-02-404	*94.1*	*93.7*	*92.6*	*94.5*	*92.3*
02-02-401	*95.3*	*95.2*	*93.9*	*95.7*	*94*
02-02-319	*92.8*	*91.7*	*91*	*93.4*	*90*

	02-02-505	02-02-504	02-02-412	02-02-404	02-02-401	02-02-319
W44	38.6	38.6	38.5	37.9	38.4	37.8
W41	75.2	75	74.7	74	74.5	72.8
W38	36.7	36.8	36.6	36.6	36.3	35.4
W35	39.9	40	39.9	39.5	39.4	39.1
W28	38.2	38.2	38.1	37.6	37.8	37.7
W15	*87.7*	*87*	86.5	*87.5*	86.8	83.3
W12	35.1	34.9	34.7	34.9	34.7	34.9
S49	31.1	31	31	30.4	30.7	30.3
S45	76.8	77.3	76.7	75.6	75.8	74
S44	40.9	40.9	41.1	40.6	40.5	39.8
S41	81.2	80.4	80.8	79.6	79.9	77.3
S39	38.5	39.3	38.5	37.6	37.8	37.8
S36	40.7	40.6	40.7	40.2	40.1	39.7
S35	30.2	29.7	30.2	29.4	29.6	28.8
S34	*91.5*	*91.6*	*92*	*89.4*	*91*	*89.3*
S32	39.7	39.7	39.7	39.5	39.3	38.7
S31	39	39.4	39	39	38.6	38.1
S28	83.4	84.5	82.7	81.8	82.3	78.4
S27	36.4	36.5	35.8	35.6	35.5	36.4
S24	*89*	*89.8*	*89.5*	86.8	*88.3*	86.8
S22	41.5	41.1	41.4	40.2	40.5	40.2
S21	*96.5*	*95.3*	*95.9*	*93.8*	*93.9*	*91.5*
S20	*95.2*	*94.6*	*95.3*	*94.4*	*95.6*	*90.5*
S19	*93.6*	*95.1*	*93.3*	*90.7*	*91.9*	*90.6*
S18	*93.7*	*95.2*	*93.3*	*90.8*	*92*	*90.6*
S15	*89.4*	*90.1*	*89.9*	*87.1*	*88.5*	*87*
S14	*89.3*	*90*	*89.6*	*87*	*88.3*	86.7
S13	*88.8*	*89.5*	*89.5*	*87.3*	*87.8*	86.5
S12	*88.6*	*89.3*	*89.3*	*87.3*	*87.7*	86.3
S11	*88*	*87.1*	86.9	85.6	86.9	83.4
S10	*87.6*	86.2	86.7	85.7	86.2	83.3
S9	*87.8*	*87.2*	*87.7*	85.4	*87.1*	85.3
S8	*93.5*	*93.2*	*93.3*	*91*	*92.5*	*89.9*
S7	*92.6*	*92.2*	*92.3*	*90*	*91.6*	*89*
S2	*89.8*	*89.6*	*90.2*	*88*	*89.3*	*87.8*
S1	80	79.7	80.2	78.8	80	78.1
06-02-503	*97.9*	*96.3*	*97.3*	*94.1*	*95.3*	*92.8*
06-02-204	*96.6*	*95.9*	*95.8*	*93.7*	*95.2*	*91.7*
06-02-121	*95.9*	*94.7*	*95.8*	*92.6*	*93.9*	*91*
02-02-507	99.1	*96.3*	*97.3*	*94.5*	*95.7*	*93.4*
02-02-506	*95.1*	*96.5*	*94.1*	*92.3*	*94*	*90*
02-02-505	100	*96.6*	*97.6*	*94.8*	*96.2*	*93.7*
02-02-504	*96.6*	100	*95.5*	*93.3*	*94.7*	*91.3*
02-02-412	*97.6*	*95.5*	100	*94.4*	*95.6*	*92.7*
02-02-404	*94.8*	*93.3*	*94.4*	100	*96*	*90.4*
02-02-401	*96.2*	*94.7*	*95.6*	*96*	100	*91.2*
02-02-319	*93.7*	*91.3*	*92.7*	*90.4*	*91.2*	100

Chapter VI

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