Influence of brewing temperature and brewing period on the microbial kinetics in herbal infusions

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Introduction

Herbal infusions are traditionally prepared by brewing with boiling or nearly boiling water to extract the ingredients within a short period and to consume the beverage while still hot. Producers continue to stipulate preparation with boiling water and brewing periods of up to 10 minutes in order to develop the full flavour and to reduce the microbial load of the product. The convenience, however, to prepare infusions with water of lower temperatures, for example from the tap, has apparently changed the preparation practices amongst some consumers. Thus, in a case control study performed during an outbreak investigation by the German Federal Health Institute (Robert Koch Institute Berlin) 15 – 33 % of interviewed mothers in Germany admitted that they prepared herbal drinks for babies and infants using water below boiling temperature (ROBERT KOCH INSTITUT, 2004). This indicates a change of the consumers' practice.

Herbs, like spices and other dried plant products, naturally contain high numbers of bacterial and fungal microorganisms. Depending on the type of product, total bacterial counts may vary between 10^5 and10^8 bacteria per gram, consisting mainly of spore-forming bacilli, yeasts and moulds with 10^2 – 10^6 per gram and *Escherichia coli* with 10^2 - 10^4 organisms per gram (LEIMBECK, 1987; KOLB, 1999). An investigation was therefore conducted to examine the kinetics of bacteria and fungi in herbal infusions under different brewing temperatures and brewing periods to evaluate potential hazards associated with herbal infusions that are prepared with water of lower temperature.

Material and methods

Brewing of herbal infusions

Sterile 250 ml cups were filled with 200 ml of sterilized water (121 °C, 20 min). These were placed in a water bath until they reached the appropriate temperature. The usual amount of herbal product in a tea bag (i.e. 1.5 g camomile, 2 g mint, rose hip or hibiscus, or 3 g fruit tea) was weighed out in a tea bag and placed into the water for the respective brewing period. Before removal, the bag was agitated for better release of the herbal extracts. Samples were taken immediately for analysis. For storage tests, one cup brewed at 90 °C was placed in crushed ice for rapid cooling to room temperature and the other was cooled to room temperature under ambient conditions.

Bacterial and fungal counts

Total viable counts (TVC). To determine the natural levels of microorganisms present in the dried product, samples of 10 g of dried herbal product were homogenized in a stomacher for 60 sec with 90 ml sterile saline diluent containing 0.1 % (w/v) peptone (trypsin-digested casein). The homogenate was serially diluted 1:10 and a 0.1 ml aliquot of each dilution was spread onto plate count agar (Merck, Darmstadt, Germany), followed by incubation at 30 °C for 72 h.

For the determination of bacteria present in the infusions, the beverage was serially diluted 1:10 and 0.1 ml volumes of each dilution were transferred to petri dishes and mixed with 10 ml of liquid plate count agar (50 °C). After solidification of the agar the plates were incubated for 72 h at 30 °C; enumeration of colony forming units was performed using a 6-fold magnification lens. All tests were performed in triplicate.

Bacillus (B.) cereus. 0.1 ml volumes were plated on cereus selective agar according to Mossel (CSM; Merck, Darmstadt, Germany). After 24 h at 37 °C colonies with the typical appearance of *B. cereus* (red-violet colonies with a white halo indicating lecithinase activity) were enumerated. One typical colony of each positive plate was confirmed as *B. cereus* by a set of biochemical tests (positive reactions for haemolysis on sheep blood agar, anaerobic utilization of glucose, motility, lecithinase activity, and lack of fermentation of mannitol; RHODEHAMEL and HARMON, 1998).

Enterobacteriaceae. 0.1 ml volumes were plated onto violet red-blood-dextrose agar (VRBD, Merck, Darmstadt, Germany). Plates were incubated anaerobically for 48 h at 30 °C. Gram-negative, oxidase-negative, glucose-fermenting colonies with diameters >1 mm were counted as *Enterobacteriaceae*. For determination of *E. coli*, 0.1 ml volumes were inoculated into laurylsulfate broth tubes (3 tubes per dilution, MPN) and incubated for 48 h at 37 °C. If gas was produced, subcultures were made in one tube of lactose and one of tryptophane broth (indole production). The cultures were incubated at 44 °C for 24 h, and a positive result in both test tubes was taken as confirmation of *E. coli*.

Enterococcus species. 0.1 ml volumes were plated onto citrate azide-Tween®-carbonate agar (CATC, Merck, Darmstadt, Germany). Plates were incubated for 24 h at 37 °C followed by another 24 h at room temperature. Red, catalase-negative colonies were enumerated as enterococci.

Yeasts and moulds. 0.1 ml volumes were plated onto yeast extract glucose chloramphenicol agar (YGC, Merck, Darmstadt, Germany). Plates were incubated aerobically for 96 h at 25 °C, and yeast and mould colonies were enumerated and recorded separately.

Results

Product analysis

The numbers of microorganisms (cfu/g) determined in the dry herbs are summarized in Table 1. The counts varied not only among the five types of herbal products, but also between different lots, as determined for camomile and mint.

Brewing temperature

Examination of the kinetics of *Enterobacteriaceae*, yeasts and moulds in camomile and mint infusions at 20, 50, 70, 90, and 100 °C showed a complete inactivation of these organisms at 70 °C, regardless of the brewing periods (Figs. 1 and 2). In contrast, total viable counts revealed a two-fold increase of the organisms between 50 and 70 °C in camomile, and between 70 and 90 °C in mint infusion, respectively. This suggested a heat activation of spores present in the product. In order to validate this observation, a further series of tests was performed on camomile, mint, hibiscus, rose hip, and fruit infusions at temperatures of 20, 50, 60, 70, 80, 90, and 100 °C. The rose hip infusion contained too low a number of organisms to

Table 1: **Microbial counts (cfu/g) in dried herbal products [median of 6 tests]**

Product		TVC[b]	*Enterobacteriaceae*	*E. coli*	*Enterococcus* spp.	*B. cereus*	Yeasts	Moulds
Camomile Fine Cut	a)[a]	1.4×10^7	7.8×10^3	460	n.d.	2.0×10^4	1.8×10^4	6.0×10^3
	b)[a]	6×10^4	8.6×10^3	n.d.[c]	105	150	n.d.	< 10
Mint Fine Cut	a)	1.3×10^7	3.3×10^3	1.1×10^3	n.d.	3.0×10^3	6.6×10^6	1.2×10^4
	b)	5.5×10^6	2.4×10^4	n.d.	1.1×10^3	250	n.d.	< 10
Rose Hip Fine Cut		30	< 10	< 10.	< 10	< 10	n.d.	< 10
Hibiscus Fine Cut		5.7×10^3	< 10	< 10	< 10	52	n.d.	550
Fruit Tee Fine Cut		4.2×10^3	< 10	< 10	< 10	< 10	n.d.	< 10

[a] Samples of two different lots were examined, [b] Total aerobic viable counts, [c] Not determined

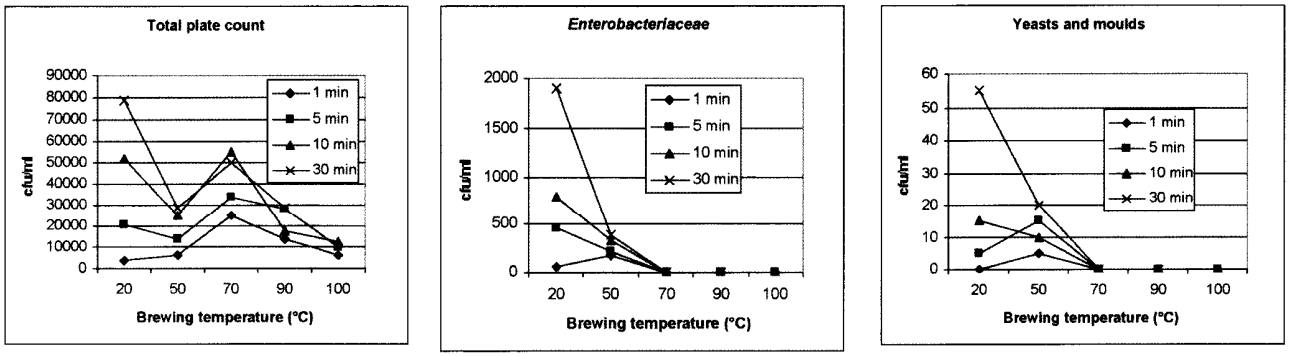

Fig. 1: Microbial counts (TVC, *Enterobacteriaceae*, yeasts and moulds, cfu/ml fluid) in camomile infusions after different brewing periods and temperatures.

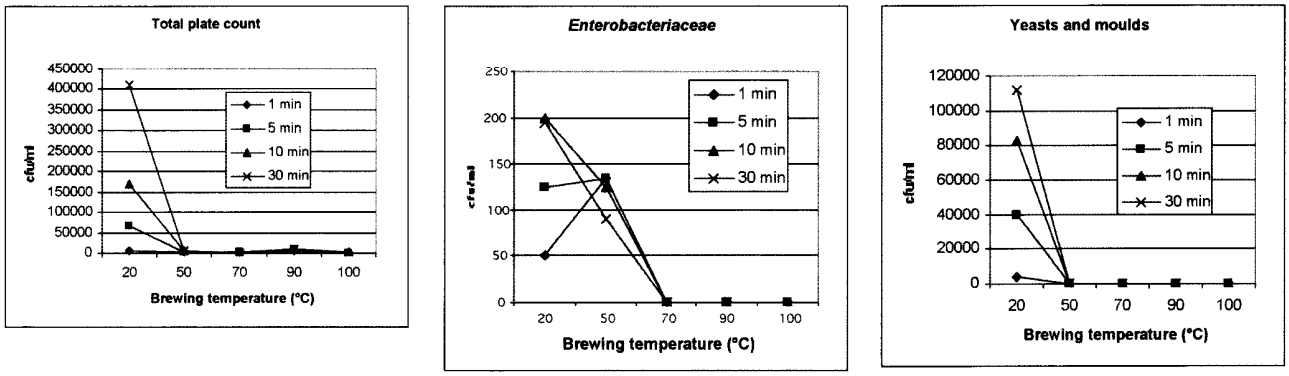

Fig. 2: Microbial counts (TVC, *Enterobacteriaceae*, yeasts and moulds, cfu/ml fluid) in mint infusions after different brewing periods and temperatures.

establish the microbial kinetics (data not shown). The remaining four products revealed a second peak of bacterial counts between 60 °C and 80 °C (mint infusion 60 °C; fruit infusion 70 °C; hibiscus and camomile infusions 80 °C). The early peak seen at 60 °C in mint infusion might be explained by the markedly higher initial bacterial load of this sample when compared to the remaining three products. The results are summarized in Figures 3 to 6.

Brewing period

Figures 3 to 6 indicate prolonged brewing consistently liberated higher numbers of microorganisms. Generally, the lower the brewing temperature, the more pronounced was the difference in bacterial counts. At temperatures above 80 °C the heat-related reduction of organisms resulted in counts having a similar level independent of the brewing period.

Extended storage of infusions at room temperature

Camomile and mint infusions were brewed for 1, 5, 10, and 30 min at 20 and 90 °C followed by storage at room temperature for 12 and 36 h (Tables 2 and 3). In both types of infusion, the TVC increased by 2–3 logs between 12 and 36 h of storage at room temperature. Depending upon the brewing periods, a gradual increase from 10^4 to 10^6 organisms per ml at 1 min brewing and 12 h storage time to a maximum of 10^8 per ml after 30 min brewing and 36 h storage was observed.

In camomile infusions, *Enterobacteriaceae* yielded 2-3 log higher numbers after 36 h storage at room temperature, when compared to 12 h. Different brewing periods resulted in an increase from 420 organisms per ml after 1 min brewing and 12 h storage to 8.3 x 10^6 organisms per ml after 30 min brewing and 36 h storage (Table 2). Similar results were obtained in mint infusions. Here the differences between 12 and 36 h storage averaged 1-2 logs, and the increase from 1 min brewing and 12 h storage to 30 min brewing and 36 h storage was from 610 to 3.5 x 10^7 *Enterobacteriaceae* per ml (Table 3).

In both types of infusions stored at room temperature the numbers of *E. coli* increased by 5 logs between 1 min brewing with 12 h storage, and 30 min brewing followed by 36 h storage. Maximum numbers of 4.6 x 10^6 per ml in camomile and 4.3 x 10^5 per ml in mint infusions were reached (Tables 2 and 3).

Yeast and mould counts were low in camomile infusions with numbers of < 10 to 10^2 per ml (Table 2). In contrast, in mint infusions, yeast numbers between 10^3 and 10^5 per ml were observed without a clear tendency to increase after different brewing and storage periods. Moulds, on the other hand, increased from 50 to > 10^3 with prolonged brewing and storage times (Table 3).

Brewing at 90 °C completely eliminated *Enterobacteriaceae*, *E. coli*, yeasts and moulds, regardless of the brewing periods. Slow or fast cooling of the infusions to room temperature did not influence the subsequent propagation of the surviving microorganisms. Extension of the storage period from 12 to 36 h increased the TVC by 1-2 logs. The average counts were approximately 10^4 per ml after 12 h, increasing to 10^6 per ml in camomile and to 5 x 10^5 per ml in mint infusions (Tables 2 and 3).

Different lots of camomile and mint markedly varied in their *B. cereus* content (Table 1), which resulted in about 10^3 cfu/ml (lot 1) and 10 cfu/ml (lot 2) infusion brewed at 100 °C. Their number increased by 1 log during storage for 24 hours. For the other three herbal infusions the *B. cereus* counts were below 10 cfu/ml.

The pH of infusions

The pH of infusions prepared from camomile and mint ranged from 8.3 to 7.2, while rose hip, hibiscus and fruit infusions were more acidic giving a pH range from 7.6 - 4.8, 3.5 - 2.8, and 4.7 - 2.7, respectively (data not shown).

Discussion

The results confirmed that different types of dried herbal products vary considerably with regard to their natural microbial load (Table 1). Leafy products such as camomile and mint contain significantly higher numbers of microorganisms than non-leaf products e.g. rosehip or fruits. This may be due to their surface structure and the close proximity of leaves to the soil.

Natural factors during growth, harvest, and primary processing may influence the microbial load of non-pathogenic organisms and potential pathogens such as *B. cereus*, moulds, and *Enterobacteriaceae* including *Salmonella*, which are present in the environment. Inactivation or survival of these microorganisms are determined by the brewing temperature and the brewing period. These are key factors to ensure that these microorganisms are not present in the prepared infusion.

Growth of surviving microorganisms may be inhibited by intrinsic factors of the product such as pH and volatile oil components con-

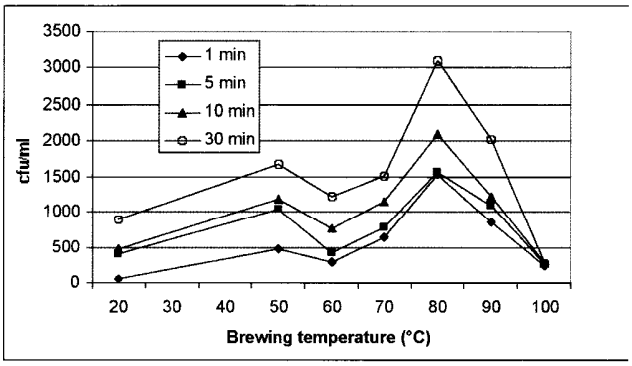

Fig. 3: Total aerobic counts (cfu/ml fluid) in camomile infusions after different brewing periods and temperatures [mean of 6 tests].

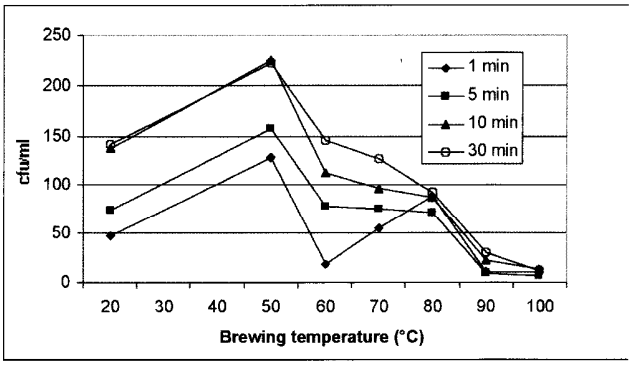

Fig. 5: Total aerobic counts (cfu/ml) in hibiscus infusions after different brewing periods and temperatures [mean of 6 tests].

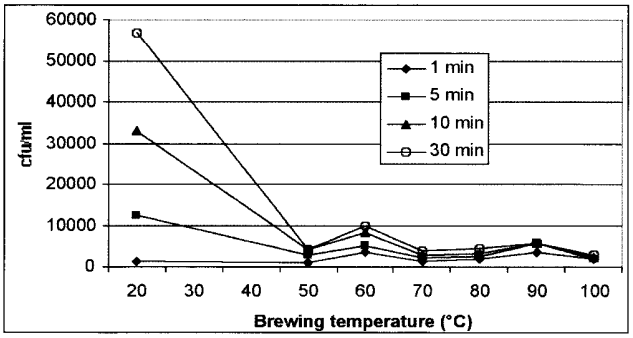

Fig. 4: Total aerobic counts (cfu/ml) in mint infusions after different brewing periods and temperatures [mean of 6 tests].

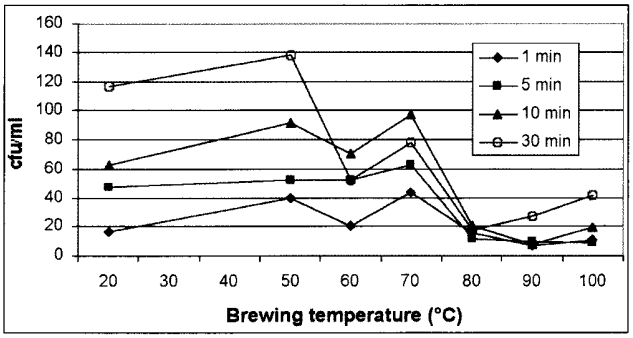

Fig. 6: Total aerobic counts (cfu/ml) in fruit infusions after different brewing periods and temperatures [mean of 6 tests].

taining antimicrobial properties, or supported by the presence of fementable carbohydrates. These factors, however, were not examined in the present study.

As has been confirmed by this study, preparation with boiling or nearly boiling water (90 °C) eliminates the vegetative microbial flora (Figs. 1 and 2; Tables 2 and 3). However, incorrect preparation of infusions by convenience or change of consumers' practice, using water of much lower temperatures or even taken from the tap, allow vegetative organisms to survive. Such misuse seems not to be unusual. In a case control study performed during an outbreak investigation by the German Federal Health Institute 15-33 % of interviewed mothers in Germany prepared herbal drinks for babies and infants with water below boiling temperature (ROBERT KOCH INSTITUT, 2004). This increases the risk that pathogens, which may be present in the product, may be transferred into the beverage.

Table 2: **Microbial counts per ml camomile infusion after 12 and 36 hours storage at room temperature, following preparation of the infusions at 20 °C and 90 °C for different periods**

Brewing temperature	Brewing period	Duration of storage	TVC	*Enterobacteriaceae*	*E. coli*	Yeasts	Moulds
20 °C	1 min	12 hours	9.3×10^3	420	48	< 10	< 10
20 °C	1 min	36 hours	8.5×10^6	4.4×10^5	4.3×10^4	50	< 10
20 °C	5 min	12 hours	7.5×10^4	4.5×10^3	280	< 10	< 10
20 °C	5 min	36 hours	3.9×10^7	1.4×10^6	2.4×10^6	20	< 10
20 °C	10 min	12 hours	4.9×10^5	1.1×10^4	1.5×10^4	40	10
20 °C	10 min	36 hours	6.0×10^7	5.4×10^6	2.4×10^6	50	< 10
20 °C	30 min	12 hours	1.2×10^6	2.9×10^4	2.4×10^3	110	20
20 °C	30 min	36 hours	1.1×10^8	8.3×10^6	4.6×10^6	90	50
90 °C	1 min	12 hours after slow cooling[a]	1.4×10^4	< 10	< 3 [b]	< 10	< 10
90 °C	1 min	36 hours after slow cooling	8.9×10^5	< 10	< 3	< 10	< 10
90 °C	1 min	12 hours after fast cooling[a]	1.0×10^4	< 10	< 3	< 10	< 10
90 °C	1 min	36 hours after fast cooling	1.2×10^6	< 10	< 3	< 10	< 10
90 °C	5 min	12 hours after slow cooling	3.8×10^4	< 10	< 3	< 10	< 10
90 °C	5 min	36 hours after slow cooling	7.8×10^5	< 10	< 3	< 10	< 10
90 °C	5 min	12 hours after fast cooling	2.8×10^4	< 10	< 3	< 10	< 10
90 °C	5 min	12 hours after fast cooling	5.4×10^5	< 10	< 3	< 10	< 10
90 °C	10 min	12 hours after slow cooling	3.5×10^4	< 10	< 3	< 10	< 10
90 °C	10 min	36 hours after slow cooling	8.9×10^5	< 10	< 3	< 10	< 10
90 °C	10 min	12 hours after fast cooling	3.0×10^4	< 10	< 3	< 10	< 10
90 °C	10 min	36 hours after fast cooling	4.0×10^5	< 10	< 3	< 10	< 10
90 °C	30 min	12 hours after slow cooling	5.1×10^4	< 10	< 3	< 10	< 10
90 °C	30 min	36 hours after slow cooling	1.7×10^6	< 10	< 3	< 10	< 10
90 °C	30 min	12 hours after fast cooling	3.9×10^4	< 10	< 3	< 10	< 10
90 °C	30 min	36 hours after fast cooling	1.2×10^6	< 10	< 3	< 10	< 10

[a] Slow cooling, cooling down the infusion at room temperature; fast cooling, cooling down the infusion to room temperature by placing the container in crushed ice; [b] MPN enumeration

Table 3: **Microbial counts per ml mint infusion after 12 and 36 hours storage at room temperature, following preparation of the infusions at 20 °C and 90 °C for different periods**

Brewing temperature	Brewing period	Duration of storage	TVC	*Enterobacteriaceae*	*E. coli*	Yeasts	Moulds
20 °C	1 min	12 hours	1.1×10^4	610	2	2.8×10^3	50
20 °C	1 min	36 hours	1.7×10^7	2.8×10^6	7.5×10^3	2.8×10^5	1.0×10^3
20 °C	5 min	12 hours	4.7×10^5	1.8×10^5	2.4×10^3	9.1×10^4	700
20 °C	5 min	36 hours	6.2×10^7	4.9×10^6	1.5×10^6	7.0×10^4	900
20 °C	10 min	12 hours	1.0×10^6	4.6×10^5	4.3×10^3	2.1×10^5	4.0×10^3
20 °C	10 min	36 hours	7.6×10^7	1.1×10^7	9.3×10^5	1.9×10^4	?[a]
20 °C	30 min	12 hours	3.5×10^6	1.1×10^6	9.3×10^3	2.3×10^5	1.3×10^3
20 °C	30 min	36 hours	1.1×10^8	3.5×10^7	4.3×10^5	1.2×10^4	?[a]
90 °C	1 min	12 hours after slow cooling[b]	6.1×10^3	< 10	< 3[c]	< 10	< 10
90 °C	1 min	36 hours after slow cooling	4.2×10^5	< 10	< 3	< 10	< 10
90 °C	1 min	12 hours after fast cooling[b]	6.2×10^3	< 10	< 3	< 10	< 10
90 °C	1 min	36 hours after fast cooling	2.0×10^5	< 10	< 3	< 10	< 10
90 °C	5 min	12 hours after slow cooling	9.1×10^3	< 10	< 3	< 10	< 10
90 °C	5 min	36 hours after slow cooling	2.5×10^5	< 10	< 3	< 10	< 10
90 °C	5 min	12 hours after fast cooling	8.7×10^3	< 10	< 3	< 10	< 10
90 °C	5 min	12 hours after fast cooling	1.1×10^5	< 10	< 3	< 10	< 10
90 °C	10 min	12 hours after slow cooling	8.9×10^3	< 10	< 3	< 10	< 10
90 °C	10 min	36 hours after slow cooling	2.4×10^5	< 10	< 3	< 10	< 10
90 °C	10 min	12 hours after fast cooling	1.0×10^4	< 10	< 3	< 10	< 10
90 °C	10 min	36 hours after fast cooling	9.2×10^4	< 10	< 3	< 10	< 10
90 °C	30 min	12 hours after slow cooling	9.7×10^3	< 10	< 3	< 10	< 10
90 °C	30 min	36 hours after slow cooling	5.2×10^5	< 10	< 3	< 10	< 10
90 °C	30 min	12 hours after fast cooling	1.0×10^4	< 10	< 3	< 10	< 10
90 °C	30 min	36 hours after fast cooling	3.0×10^5	< 10	< 3	< 10	< 10

[a] Moulds were overgrown by bacteria and yeasts and could not be detected; [b] Slow cooling, cooling down the infusion at room temperature; fast cooling, cooling down the infusion to room temperature by placing the container in crushed ice; [c] MPN enumeration

Immunocompromised individuals, infants or the elderly may be at an increased risk of illness if beverages prepared in this way are consumed. This has been recently demonstrated by an outbreak with *Salmonella* Agona in a number of infants and young children in Germany (ROBERT KOCH INSTITUT, 2004).

Herbal infusions are also consumed as cold beverages. Such refreshments are usually prepared with boiling water and subsequently stored in the refrigerator or at ambient temperatures for longer periods to be available to the consumer when convenient. If such drinks are prepared with non-boiling water, numbers of vegetative organisms may considerably increase. In the present study, we chose 12 h and 36 h as storage periods and observed that *Enterobacteriaceae* and *E. coli*, depending on the brewing period, grew up to ca. 10^7 and 10^6 organisms per ml, respectively, if the infusion was prepared with cold instead of boiling water and subsequently kept at room temperature. Under these conditions, herbal infusions may pose an increased health risk to vulnerable groups of consumers if the product is contaminated with enteropathogenic bacteria such as *Salmonella*.

In conclusion, herbal infusions must be prepared using boiling water. These beverages may be consumed either immediately as a hot beverage or stored under refrigerated conditions for cold consumption.

Consumers should follow the preparation instructions given by the manufacturers and should be made aware of the possible consequences of improper preparation. As dried plant products including herbal infusions may be contaminated with enteropathogens such as salmonellae (BOCKEMÜHL and WOHLERS, 1984), and misuse of the consumers cannot be excluded, manufacturers must observe Good Agricultural & Hygienic Practice (GAHP; EUROPEAN HERBAL INFUSIONS ASSOCIATION, 2002) and apply stringent HACCP systems to reduce the microbial load and to ascertain the absence of enteropathogenic organisms.

Abstract

Although producers stipulate that herbal infusions should be prepared with boiling water, it is known that some consumers use water at lower temperatures. Untreated herbs and other dried plant products naturally contain a range of microorganisms which may be present in high numbers. This may include both non-pathogens and pathogens such as yeasts, moulds, *Enterobacteriaceae* and *Salmonella*. In this study, we investigated the microbial kinetics of herbal infusions after various brewing periods and temperatures.

In a first series of tests on camomile and mint infusions, *Enterobacteriaceae*, yeasts and moulds were no longer detectable at brewing temperatures above 70 °C, but the total viable count (TVC) revealed a twofold increase of the organisms at 70-90 °C. This suggested a heat activation of bacterial spores present in the product. In a second series of tests on camomile, mint, hibiscus, and fruit infusions, again a second peak of bacterial counts was detected at brewing temperatures between 60-80 °C. Brewing periods between 1 min and 30 min showed that the longer the brewing period the higher was the number of microorganisms released into the infusion.

Extended storage of camomile and mint infusions for 12 and 36 h at room temperature was tested with infusions prepared at 20 and 90 °C, respectively. The results showed the absence of *Enterobacteriaceae*, *E. coli*, yeasts and moulds when prepared with water at 90 °C, but the TVC increased to approximately 10^4 per ml after 12 h and to 10^5-10^6 per ml after 36 h storage. When prepared with water at 20 °C the TVC increased from 10^4 organisms per ml after 12 h to 10^8 per ml after 36 h storage at ambient temperature. Under the same preparation conditions *Enterobacteriaceae* and *E. coli* grew to 10^5-10^7 organisms per ml after 36 h storage. Growth of yeasts and moulds was observed at 20 °C. However, there was no clear correlation between the brewing period and storage conditions on yeast and mould counts.

This study confirms that preparation of herbal infusions with boiling water will give a safe beverage which should be consumed after preparation or stored in the refrigerator. This verifies the manufacturers' instructions to use boiling water when preparing herbal infusions. Consumer awareness of the possible consequences of misuse must be raised. Producers have to observe Good Agricultural & Hygienic Practice and apply stringent HACCP systems to reduce the microbial load and to ascertain the absence of enteropathogens.

Zusammenfassung

Obwohl die Hersteller darauf hinweisen, dass Tee-ähnliche Getränke mit kochendem Wasser zuzubereiten sind, hat sich das Verbraucherverhalten in sofern geändert, dass nicht wenige Konsumenten hierfür Wasser mit geringerer Temperatur verwenden. Unbehandelte Pflanzen oder getrocknete Pflanzenprodukte sind mit verschiedensten Mikroorganismen behaftet, die in großer Zahl vorhanden sein können. Darunter finden sich sowohl apathogene als auch pathogene Mikroorganismen wie Hefen, Schimmelpilze, *Enterobacteriaceae* und Salmonellen. In dieser Studie untersuchten wir den Einfluss verschiedener Brühzeiten und Brühtemperaturen auf die Kinetik der mikrobiellen Flora.

In einer ersten Testserie mit Kamillen- und Minzetee waren *Enterobacteriaceae*, Hefen und Schimmelpilze ab einer Brühtemperatur von 70 °C nicht mehr nachzuweisen, während sich die Gesamtkeimzahl bei Brühtemperaturen zwischen 70 und 90 °C verdoppelte. Dies ließ auf eine Hitzeaktivierung von Bakteriensporen schließen, die in den Proben nachzuweisen waren. In einer zweiten Testserie mit Kamille, Minze, Hibiskus und Früchtetee wurde wiederum ein zweiter Keimzahlgipfel bei Brühtemperaturen zwischen 60 und 80 °C gemessen. Unterschiedliche Brühzeiten zwischen 1 und 30 Minuten hatten zur Folge, dass mit zunehmender Extraktionszeit auch die Zahl der freigesetzten Mikroorganismen anstieg.

Die Auswirkung langer Standzeiten von Kamillen- und Minzetees, die mit Wasser von 20 oder 90 °C hergestellt wurden, wurde nach 12 und 36 Stunden gemessen. In den bei 90 °C gebrühten Tees wurden *Enterobacteriaceae*, *E. coli*, Hefen und Schimmel nicht mehr nachgewiesen, aber die Gesamtkeimzahl stieg auf 10^4/ml nach 12 und auf 10^5-10^6/ml nach 36 Stunden Standzeit bei Raumtemperatur. In den bei 20 °C angesetzten Tees stieg die Zahl der Mikroorganismen von 10^4/ml nach 12 auf 10^8/ml nach 36 Stunden. Unter diesen Bedingungen waren nach 36 Stunden 10^5-10^7 *Enterobacteriaceae* und *E. coli* pro ml nachzuweisen. Ein Wachstum von Hefen und Schimmelpilzen wurde zwar beobachtet, aber eine klare Korrelation zwischen Brühbedingungen und Standzeiten war nicht festzustellen.

Die Studie zeigt, dass Kräuter- und Früchtetees mit kochendem Wasser hergestellt und umgehend konsumiert oder im Kühlschrank gelagert werden müssen, um ein mikrobiell stabiles und sicheres Produkt zu erhalten. Die Verbraucher müssen für die möglichen Folgen eines abweichenden Brühverhaltens noch sensibilisiert werden. Deshalb sind auch die Hersteller angehalten, „Good Agricultural & Hygienic Practice" und ein stringentes HACCP-Konzept vorzuhalten, um die mikrobielle Belastung ihrer Produkte zu reduzieren und die Abwesenheit enteropathogener Mikroorganismen zu sichern.

Acknowledgements

The authors are grateful to Anne Knöchlein, Norma Welsch and Gisela Allmendinger for technical assistance and to Katie Donnelly and Stuart Warren for critical discussions. The study was supported by the European Herbal Infusions Association (EHIA).

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