OSTE - a novel polymer system developed for Lab-on-Chip

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ABSTRACT

OSTE polymer has the aim to address today's dissemination gap between successful lab-on-chip research and the healthcare setting. We have formulated and demonstrated a novel, superior, polymer system, OSTE, and its manufacturing platform, which is based on the mixture of three monomers: thiols, -enes and epoxies. The uniqueness of the OSTE approach stems from the curing in two distinct steps: after the first cure, an intermediate polymer is formed which is ideally suited for surface modifications and bonding; after the second cure we obtain an inert and robust polymer. Our vision is that OSTE has the potential to form a de-facto standard for research and development of high performance labs-on-chip in academia and industry.

Keywords: OSTE, OSTE+, OSTEmer, PDMS, Lab-on-a-chip, microfluidics, polymer micromanufacturing, MEMS, point-of-care diagnostics

1. INTRODUCTION

1.1 Introduction to microfluidic manufacturing

Most of the innovation in the field of lab-on-chip (LoC) is generated in the academic world, where extensive research is performed on manufacturing techniques, improved (bio)chemical reactions, ever better performing sensors and actuators, and novel applications. Most of these innovations are realized in high-performance materials, such as silicon or glass, or elastomers such as polydimethylsiloxane (PDMS), or in a heterogeneous combination of these material systems. Glass is the standard receptacle material in chemistry and therefore a given candidate material for reproducible miniaturized systems. Silicon and the extensive MEMS and IC manufacturing toolbox, have the intrinsic promise for a large geometrical design freedom, combined with the integration of electronic, photonic, micromechanical and fluidic functions into the same device. Specifically, most of the conceptual microfluidic device development today is done using PDMS, mainly because of its ease of use and attractive mechanical elastomeric properties. It is natural to think that excellent starting points for commercial development are the thousands of conceptual devices already developed by the scientific community. Unfortunately, the vast majority of these are unsuitable for product development. While silicon and glass enable devices with very advanced features, they are typically too expensive to fabricate given that the normal size of LoCs are several cm³ and that cost targets are typically a few Euros. For most applications a prototype conceptual device based on PDMS needs complete redevelopment to realize a commercially viable thermoplastic device, because: i) the elastomeric properties are inadequate for many commercial products that need invariable surface chemistry and mechanical strength, and ii) the PDMS fabrication processes are difficult to scale up because they contain fabrication steps, such as slow thermal curing and manual handling of easily strained device layers, that are incompatible with medium or large scale manufacturing.

Currently the only alternative for commercial production of LoCs is microinjection molding or hot embossing using thermoplastics such as COC and PMMA.^{1,2} Moving from conceptual tests in silicon, glass or PDMS to (mass) production requires a switch of technology since materials properties differ between research devices and those made from thermoplastics. This switch necessitates at least an entire redesign and re-evaluation cycle before a viable prototype is reached, constituting a gap, in economic terms, between research and commercial devices. Currently the only alternative for prototyping is using the commercial scale molding or embossing methods. However, these techniques are expensive and suffer from slow mold fabrication, which renders most LoC device applications economically unviable, unless the end market demands millions of devices per annum or allows for device prices in the several tens of Euros range.

To meet demands on portability and function, the facile integration of functionalities, such as surface (bio)chemistry, heterogeneous sensor materials, or elastomeric fluidic actuators, are required. In particular, durable, uniform and repeatable surface modifications, and bonding to biofunctionalized surfaces have proven difficult, both of which are necessary to ensure leak free devices with excellent performance and long shelf life. Because fundamental scaling laws lead to surface properties dominating the behavior of microfluidic devices, properly engineered and reproducible surface properties are of the highest importance. Surface modifications typically require surface activation, since most thermoplastics lack reactive surface chemistries. The most commonly used method for surface activation is oxygen plasma, which produces highly reactive chemical groups that readily form covalent bonds to molecules that are subsequently used as anchors for further modifications. While relatively uncomplicated to perform, plasma transport is limited in hard to reach areas, e.g. in the bottom of channels and below undercuts, which tends to result in uneven or inadequate surface modification. Hence, advanced devices are difficult to properly surface coat, which renders them unreliable and decreases device yield.

Bonding of individual device layers to form a sealed device is a problem that all above described LoC manufacturing technologies have in common. Today there exists no uncomplicated and cost-effective manner for bonding of the microfluidic structures after (bio)functionalization, because the biochemistry typically does not withstand elevated temperatures, solvents or plasma treatment. The most versatile strategy for bonding microfluidic device parts is gluing.^{3,4,5} While general and biocompatible, great care must be exerted to ensure that liquid glue does not block microfluidic features,^{6,7} which tends to add complexity and cost to the back-end processing. Alternative thermoplastic polymer bonding techniques function by polymer chain diffusion across a bond interface to form a monolithic structure. Typically heat or solvents combined with high pressures are needed for good results. However, interdiffusion requires very similar polymeric materials in the joined parts, since the success ultimately relies on the solubility of one type of polymer into another. Thus, good results are reached only when the materials are of the same kind, e.g. cyclic olefin copolymer (COC) bonded to COC, which limits the range of designs possible since elastomeric materials, for pumps and valves, are incompatible with COC. Moreover, when solvents are used to soften the interface, remaining solvent in the joint will leach during storage of the device, risking denaturing of on-board capture probes and may initiate stress cracks (crazing). Thus, today, no satisfactory solution for rapid prototyping of commercially viable low to medium production volume devices is available. In addition to the lack of a viable prototyping process, current commercial devices are hampered by limited device complexity and a lack of integrated functional features.

1.2 Introduction to thiol-ene chemistry

Hoyle and Bowman⁸ review thiol-ene chemistry, and mention knowledge of thiol-ene chemistry already in 1905⁹. NOA63 (Norland Optical Adhesive, Norland Products Inc., NJ) is a thiol-ene glue used since 2007 for microfluidic structure replication.¹⁰ However, NOA is expensive and not adapted for layer bonding or the tuning of the mechanical and surface chemistry properties. Off-stoichiometric mixtures of thiol-enes were first mentioned in a 1972 patent¹¹, but did not find their way into applications until our team re-invented them in search for a solution to the manufacturing problems in labs-on-chip. Since the introduction of PDMS soft lithography (1994)¹², and thick SU8 photoresist (1997)¹³, no fundamental material developments have occurred in the academic field of lab-on-chip, until our team introduced the off-stoichiometry thiol-ene (OSTE) polymer system in Oct. 2010.¹⁴

2. OFF-STOICHIOMETRY THIOL-ENE (OSTE)

2.1 Off-Stoichiometry Thiol-Ene (OSTE)

OSTE is a new polymer platform for fabrication of microfluidic devices, and the first polymer system designed specifically for the field of lab-on-chip. We demonstrated its potential for tuneable mechanical properties¹⁵, tuneable surface chemistry^{16,17}, adhesive-free room temperature bonding¹⁸, one-step photopatterning and molding for integrated vias¹⁹, and high resolution, high aspect-ratio microstructuring using an enhanced stoichiometry-dependent photolithography mechanism²⁰, and packaging of biosensors²⁶ (Fig. 1, Table 1). By Dec. 2013, OSTE-polymers were already in use in > 25 labs worldwide. Lafleur et al. reported protein surface patterning on different OSTE materials using the available thiol surface anchors²¹, Mongkhontreerat et al. fabricated tuneable cross-linked functional hydrogel networks, patterned and postfunctionalized to have superhydrophobic properties²², XX et al used the temperature dependent material stiffness for improved neural probe design²³, and Simon et al. formed gold layers using gold nanoparticles in OSTE devices for use as electrodes²⁴.

However, whereas OSTE in many aspects already exceeds the performance of other polymers for LoC manufacturing, it

exhibits crucial drawbacks including: 1) to enable bonding of OSTE layers to each other, two different types of OSTE materials with their corresponding specific surface chemistry are required, one material with -ene functional groups and one material with thiol functional groups; 2) the native OSTE surface has limited hydrophilicity, necessitating surface functionalization to ensure proper fluidic behavior; 3) leaching of unreacted monomers; and 4) limited high temperature performance, i.e. low glass transition temperature, T_g , which prevents LoCs operating at elevated temperatures typically needed in DNA assays (Table 1).

Table 1. Performance of OSTE and OSTE+

	single cure, binary monomer, OSTE	Dual cure, ternary monomer, OSTE+
Surface properties	Full design freedom in terms of surface chemistry during manufacturing. Need for surface passivation/blocking post manufacturing.	Full design freedom in terms of surface chemistry during manufacturing after first cure. Inert surface after second cure.
Mechanical properties	Stiffness tuneable: 10 MPa < E < 2000 MPa demonstrated. Limited temperature performance: $T_g < 70^\circ\text{C}$ (not suitable for, e.g., PCR processing). Monomer leaching.	Stiffness tuneable for manufacturing (after first cure). Stiffness tuneable for final material properties (after second cure): 2.1 MPa < E < 5000 MPa (prel. results). High temperature stable: $T_g > 115^\circ\text{C}$ (prel. results). No monomer leaching (<0.1% weight loss in chloroform 24 hrs).
Bonding	Low-temperature, biocompatible, adhesive-free bonding Bonding limited to thiol, ene or gold surface chemistries, and requires layers with different surface properties	Low-temperature, biocompatible, adhesive-free bonding Epoxy surface chemistry allows bonding to most surfaces, including those without active chemical groups

Fig. 1. Preliminary OSTE demonstrations from our lab. Left to right, top to bottom: tuneable bulk stiffness¹⁵, photopatternable hydrophilicity/ hydrophobicity¹⁶, microstructuring through direct lithography²⁰; packaged protein and DNA arrays²⁵, QCM sensor packaging²⁶, wafer-level integration¹⁸, microfluidic device manufacturing¹⁵.

2.2 Off-Stoichiometry Thiol–Ene–ePoxy (OSTE+)

To address the limitations of OSTE, we developed a novel, superior, *ternary, dual-cure*, thiol-ene-epoxy (OSTE+) polymer system. OSTE+ addresses the remaining unresolved limitations of OSTE (Table 1) and is potentially ideal for both rapid prototyping and mass fabrication of advanced LoC devices. The OSTE+ platform consists of three different monomers that are polymerized on demand in two distinct steps: the first step produces an intermediate, soft, solid material with surface properties ideal for bio/chemical modification and bonding; the second polymerization step results in inert polymers with properties similar to advanced engineering plastics. Briefly, the platform is a mixture of thiols, enes and epoxies, a radical initiator and an anionic initiator. The basic premise of the ternary monomer formulation is the ability to fully react one monomer functional group, while leaving a predetermined amount of a second monomer functional group unreacted in a first radically mediated curing step (Fig. 2). This relies on having a very rapid cross-reaction between the thiol and ene monomers while simultaneously having very low monomer self-reactivity, i.e. little or

no homopolymerisation. The material after the first step contains an excess of highly reactive thiol and epoxy groups that are ideal for: a) further surface modifications; and b) low- temperature glue free bonding to a multitude of substrates via the epoxy functional group. Upon the second separately initiated anionic polymerization of the thiol and epoxy functional groups, a highly cross-linked and solvent resistant polymer is formed.

OSTE+ is truly a platform technology. We recently started verifying (since mid 2012, partly unpublished) that the developed materials feature a well-controlled polymer chemistry and modularity, enabling the OSTE+ system to reach most of the crucial technical challenges, including:

- **Tuneable mechanical properties:** soft and rubbery materials (Young's modulus <2.5 MPa, i.e. similar to PDMS) are formulated with the same ease as very stiff materials (Young's modulus >3 GPa, i.e. similar to PMMA). Recently we demonstrated microfluidic chips combining stiff OSTE+ for structural integrity and rubbery OSTE+ for pneumatic valves and pumps²⁷. This is possible since changing the crosslink density, the relative ratio between thiol-ene and thiol-epoxy and the stiffness of individual monomer cores all affect the final properties of the polymer in a controllable manner.
- **Tuneable surface properties:** the tested materials feature well-controlled polymer chemistry and modularity, allowing permanent surface modifications of specific regions. We have shown durable uv-grafting protocols for modifying the wetting properties of OSTE+ between 20° and 110° with preserved bonding capability²⁸.
- **Adhesive-free, room-temperature, bonding:** we demonstrated adhesive-free RT bonding of OSTE+ to a multitude of substrates, including glass, Al, Si, plastics, and itself^{29,30,31}. Using the vast OSTE+ toolbox, it is now possible to: seamlessly integrate fragile silicon and glass components without using any liquid glue; fabricate devices with soft and rubbery portions integrated with hard and temperature resistant regions; and bond biofunctionalised components. Furthermore, due to the robust and simple bonding and surface modification schemes, this can be performed in a rapid prototyping environment.
- **Biocompatibility:** OSTE+ has the same biocompatibility as polystyrene and polycarbonate (stem cell incubation for 4 weeks resulted in cell morphology the same as in standard materials). Parts of cell compatibility results are published by Herranz et al.³²
- **UV-curing and photopatterning:** the OSTE+ system can be casted and photopatterned in one step, for example to directly define fluidic ports³³. Recently, we have found that the off-stoichiometric composition leads to an entirely novel photocuring mechanism, hence allowing OSTE+ to behave as an excellent photoresist, allowing high aspect ratios (>10:1) even at very thick layers (>2 mm). Together with a built-in epoxy glue that can be used for a subsequent bonding step, OSTE+, is the first thick layer photopatternable liquid prepolymer to enter the MEMS and LoC field since the introduction of SU8.
- **Ease of manufacturing:** basic material and machining processes are versatile, inexpensive and closely resemble the well-accepted standard soft lithography process in a rapid prototyping environment (demonstrator manufacturing within tens of minutes) and potentially allow for direct medium-scale volume manufacturing, which is important for a viable prototyping system.

3. CONCLUSIONS

These preliminary results support that the OSTE and OSTE+ systems can form the base for a consistent LoC manufacturing platform, and thus for a large class of translational development projects that truly realise the promises of microfluidics in the healthcare setting.

Fig. 2. Unique room temperature two-step (UV) curing mechanism of OSTE+.

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